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A STUDY OF LOW-LEVEL LASER RETINAL DAMAGE(U) JOHNS

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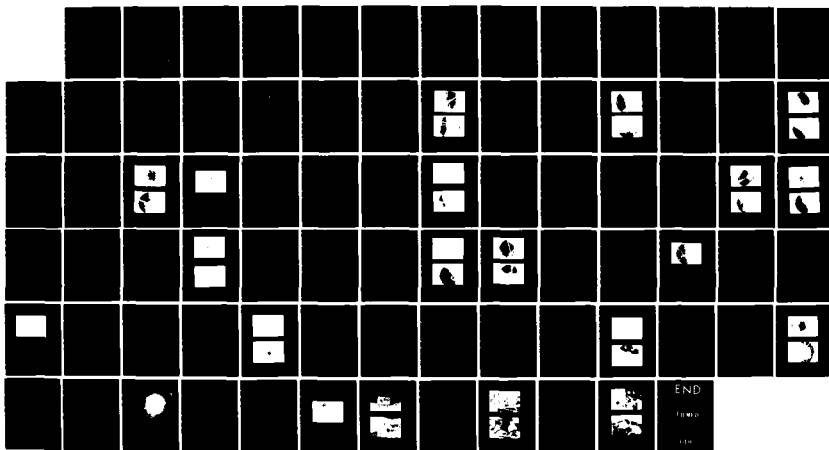
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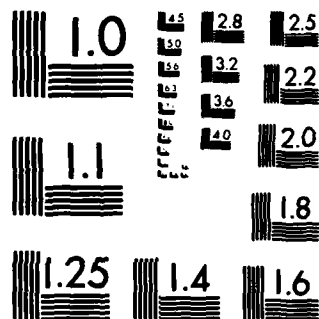
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ANNUAL
PROGRESS REPORT

for

U.S. ARMY MEDICAL RESEARCH AND DEVELOPMENT COMMAND
Fort Detrick, Frederick, MD 21701

from

RC-RCS-044
The Johns Hopkins University
Applied Physics Laboratory
Johns Hopkins Road
Laurel, MD 20707

on

A STUDY OF LOW-LEVEL LASER RETINAL DAMAGE

Approved for public release; distribution unlimited.

B. F. Hochheimer

January 15, 1985

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The findings in this report are not to be construed as an official Department of the Army position unless so designated by other authorized documents.

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FOREWORD

In conducting the research described in this report, the investigator(s) adhered to the "Guide for Laboratory Animal Facilities and Care," as promulgated by the Committee on the Guide for Laboratory Animal, Resources, National Academy of Sciences-National Research Council.

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Introduction

The objective of this program is to obtain the diffuse retinal reflectivity from a large variety of animal species.

Methods

The optical system used to record the retinal spectral reflectivity is shown in Fig. 1. The Zeiss Fundus Camera has been modified by replacing the optics in the input light path with high quality photographic objectives and a fiber optics light source. With this modification a very well-defined image of the fiber optic source can be placed on the retina. The light return path has been modified by replacing the optics, after the mirror with the central hole, with an enlarging lens, a lens stop to remove reflected light, a movable variable size retinal image aperture that limits the retinal area to only the illuminated area, a rotating interference monochrometer and a photomultiplier detector. The photomultiplier is in a coolable housing to reduce dark current noise.

This system is designed to sample only light diffusely reflected from the retina. Fig. 2 illustrates the illumination beam path and the measuring beam path inside of the eye. There is very little overlap, so that back scattered light is not detected.

An artificial eye, with a Kodak White Reflectance Standard as an artificial retina, is used as a reference. The artificial eye has a focal length of 25 mm. This white surface has known reflectance characteristics, almost 100%, and is claimed to be perfectly diffuse. The energy from the animal retina is divided by the energy from the artificial retina (at the same

Cont'd
wavelength) to remove spectral variation in light level, instrument transmission, and detector sensitivity. Originator furnished ~~Reynolds~~
includes

21473 (Figs. 1a)
The optical transmission losses in the eye are not taken into account for any of our calculations of retinal reflectivity. This severely influences the results for wavelengths shorter than 450 nm and longer than 900 nm. Correction factor could easily be included if we knew what data to use and how to separate transmission and scattering terms.

The detector output is recorded on a Princeton Applied Research model 4202 signal averager. The stepper motor driving the interference monochromator is allowed to free run and 25 data scans are averaged. This signal averager has two data channels, one for retinal data and the other for artificial eye data. A ratio of channel one to channel two is performed automatically by the averager and the data is output on an XY recorder. With this system the data collection time is less than thirty seconds.

The results given in this report cover the wavelength range from 450 to 850 nm with a spectral resolution of approximately 17 nm. The precision is approximately $\pm 5\%$ from run to run on the same animal, same retinal position, and same day. This precision drops to $\pm 10\%$ with runs taken on different days. The relative values at different wavelengths are good to $\pm 5\%$. When a retinal reflectivity of 20% is measured the variations are not more than $20 \pm 2\%$ and the relative values are somewhat better than this. The absolute accuracy is not known.

Our previous measurements and calculations indicate a large amount of light scatter in the eye. Our experimental measurements of light scatter in

the visible spectral region agree very well with those made by other researchers who used a variety of methods. All of the previous estimates of light scatter in the eye and the determinations of retinal reflectivity are based on the assumption of a completely diffuse retina, that is, the retina is assumed to be a Lambertian surface with no specular component of reflectivity. This is the view put forth by Brindley and Willmer¹ but disputed by Weale².

The apparatus we presently use will not detect a totally specular component because of the geometric arrangement of input and collection optics. Our derivation of reflected light scatter (see 1982 progress report) in the eye assumes, as a basic assumption, that the retina is diffuse.

Weale² has measured the polarized component of light reflected from the retina and computes a value $P = p/(p+d)$ where p is the polarized component and d the non-polarized component. P varies with wavelength and has values between 0.5 to 0.7. We have also shown that the retina has properties that produces a definite pattern in polarized light overlying the retina³. Our initial interpretation of this was that the retina is diffuse but retains polarization characteristics in a manner similar to an aluminized reflecting screen used for three-dimensional slides or movies. In order to completely determine the light reflective characteristics of the retina it is necessary to resolve the question of the diffuseness of the retina and light scatter in the eye (see Fig. 3).

The precision with which our measurements of reflectivity are made are rather easily defined. The absolute accuracy, however, depends on many factors. Some of these we know, but others we may not even recognize. Much

of the variability in our data is due to animal-to-animal differences and to the fact that we measure very small areas of a retina that vary in reflectivity over this surface. Many experimental details also affect the results. The measured reflectivity of the eye will depend on the size of the eye, the retinal area illuminated and probably on the utilized area of the cornea. It will also depend on any induced polarization effects in either the illumination or measurement light paths. In most measurements of reflectivity the variation with polarization vector angle is not considered, yet beam splitters are often used which must introduce polarization effects. In our modified fundus camera we have not been able to measure any induced polarization of the input light or any polarization effects in the collection optics greater than one part in 200.

Absolute retinal reflectivity is rarely, if ever, measured directly. A reference artificial retina is used as a standard. Our first measurements of retinal reflectivity⁴ used a mixture of flat white paint and carbon black to obtain a surface with 17% reflectivity as measured with a Beckman DK2 Reflectance Spectrophotometer. This surface is somewhat specular in nature. Recently I have used an Eastman White Reflectance Standard as a reference, this surface is claimed to have a known value (almost 100%) of reflectivity, and, more importantly, to be completely diffuse. With this standard our computed retinal reflectivity values have increased almost by a factor of 2x over previous results.

I am pointing out these difficulties to indicate the difficulty in the determination of the absolute accuracy of our retinal reflectivity data.

Results

A list of the animals whose retinal reflectivity has been measured is given in Table I. All of the animals were anesthetized and their pupils dilated.

Figures 4 to 18 are spectral curves of the diffuse retinal reflectivity for the animals listed in Table I. Also shown are color retinal photographs taken with a Kowa camera and Kodachrome Film (speed 64ASA). Fig. 18A is the reflectance curves for various areas of a cynomolgus monkey retina, these areas are indicated in Fig. 18B. The other photographs show experiments being done in Wyoming in June 1984.

Discussion

The curves and photographs are self-explanatory; however, as regards the spectral reflectance curves, this data is all with reference to the diffuse reflectivity from an artificial eye with a 25 mm achromatic objective lens and a totally diffuse Kodak White Reflectance Standard "retina."

Some of the reflectance curves show values in excess of 100%. If the animal retina has a surface diffusivity different from the Kodak reference standard, this greatly influences the absolute value of the results. In addition, if the animal retinas have a total or partial diffuse reflectance value, then the length of the eye from the pupil to the retina will determine how much energy is collected in our optical system. The reference eye has a 25 mm focal length. If the animal eye is longer than this the retinal reflectance curves indicate too low a value, and if it is shorter, too high a value. If the animal retina is a totally diffuse surface, the correction

Table 1

No. of
Animals

1	Antelope
2	Mule Deer
1	Moose
2	Big Horn Sheep
2	Elk
2	Domestic Cats
3	Domestic Dogs
1	Farm Goat
2	Farm Pigs
1	Great Horned Owl
2	Pigmented Rabbits
2	Albino Rabbits
2	Pigmented Guinea Pigs
2	Albino Guinea Pigs
2	Cynomolgus Monkeys
1	Rhesus Monkey (data not included in this report)

factor is the square of the ratio of the focal length of the artificial eye to that of the eye being measured.

Although the focal lengths of either the animal eye or the artificial eye vary only slightly with wavelength, due to chromatic aberration, the diffusivity may change greatly with wavelength. Polarization effects are also wavelength dependent. The spectral reflectance curves do not take into account any transmission losses or scattering in the eye. Their effects are known to be wavelength dependent.

With these things in mind we believe our data to be reasonably precise and repeatable. However, any interpretation of this data needs to be done with great care.

Unfinished Work on this Project

- 1) A rhesus monkey recently had data taken for retinal reflectivity determinations. The reduced data will be included in the final report for this project.
- 2) In January 1985 we plan to do 2 calves, 2 burros, 2 sheep and an additional rhesus monkey. This will complete our series of animal reflectance measurements in the 450-850 nm spectral region.
- 3) Our optical system will then be modified so that data can be taken in the near infrared spectral region. An S1 photomultiplier will be used in the 800 to 1000 nm region with a cat, a dog, two rabbits and a rhesus monkey as subjects.
- 4) One rhesus monkey enucleated eye retina will be analyzed on a Perkin-Elmer UV-VIS-NIR spectrophotometer equipped with an

integrating sphere reflection attachment. Such a retina will be very different from an in-vivo retina and a direct comparison may be impossible.

- 5) Other work on this program is outlined in the accompanying proposal.

Note: Funding for this animal reflectance measurement program started in Sept. 1984 and is for one year. At this time only one-third of the year has elapsed.

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J. Physiol. London 116, 350, (1952)
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J. Physiol. London 186, 175, (1966)
3. B. F. Hochheimer and H. A. Kues
Applied Optics 21, 3811, (1982).
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J. Bio. Photo. Assn. 45, 146, (1977).

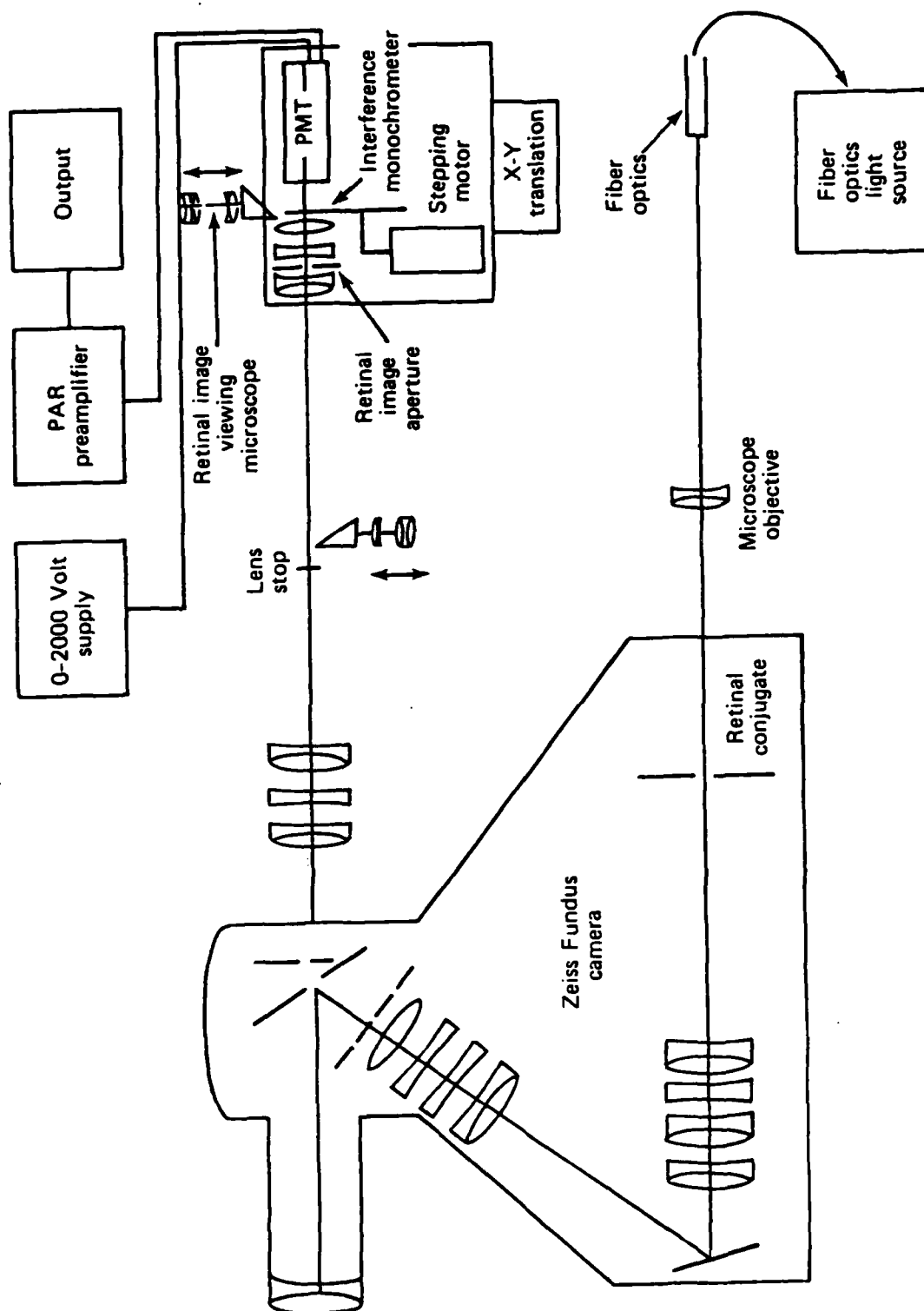


Fig. 1

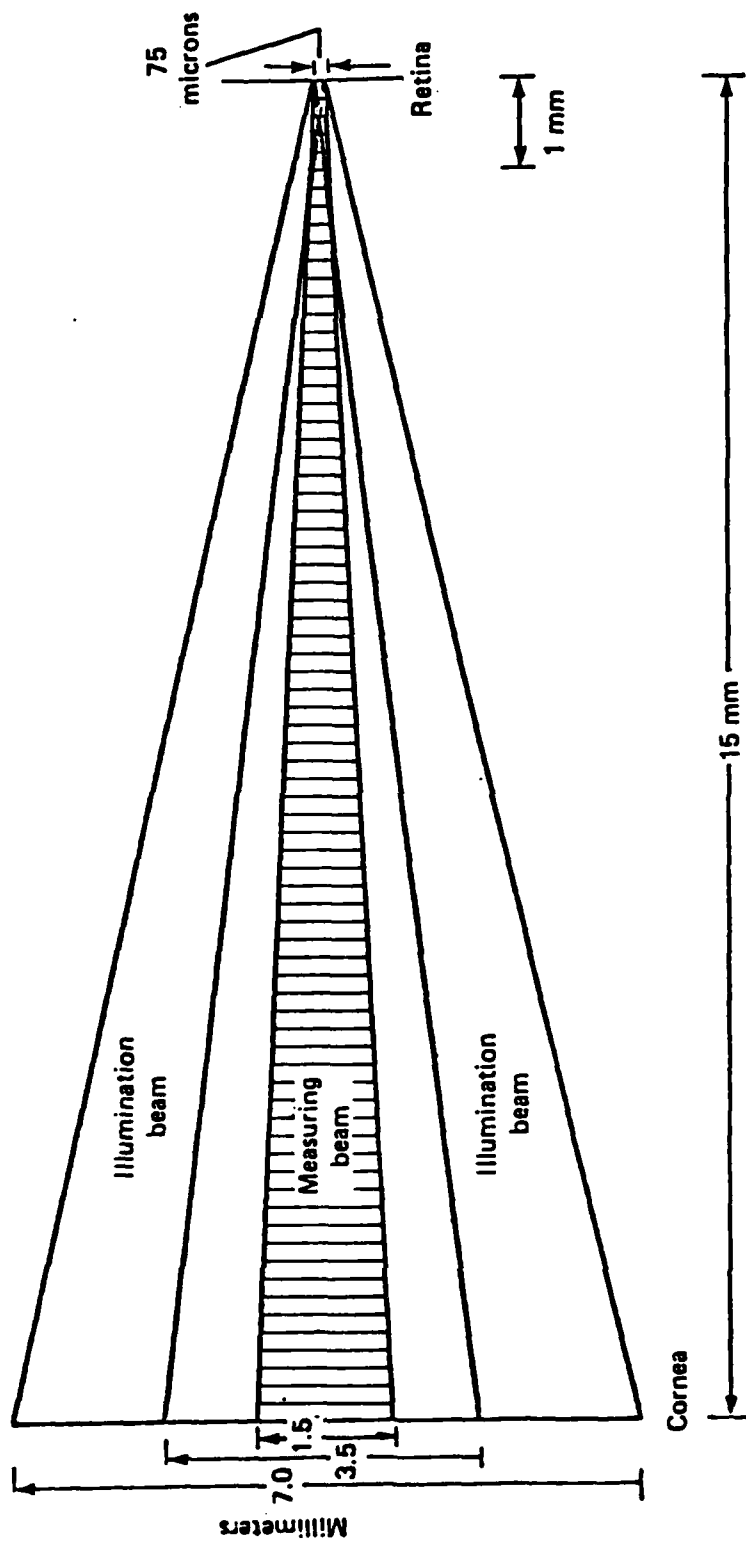
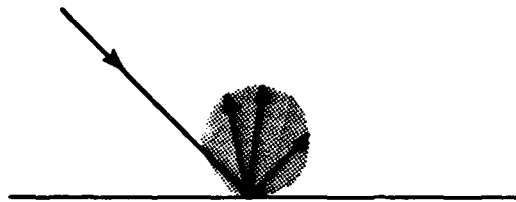
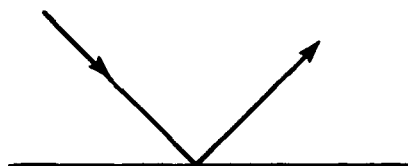


Fig. 2



A perfectly diffuse or Lambertian reflector. The brightness is independent of viewing angle.



A perfectly specular or mirror surface. Reflected light obeys Snell's law.



A partially diffuse surface.

Fig. 3

Fig. 4

Moose

In all of the following photographs the color rendition may be misleading because of overexposure. This tends to indicate a "whiter" retina than is the actual color.

The moose retina is, however, almost white with a slight bluish tinge.

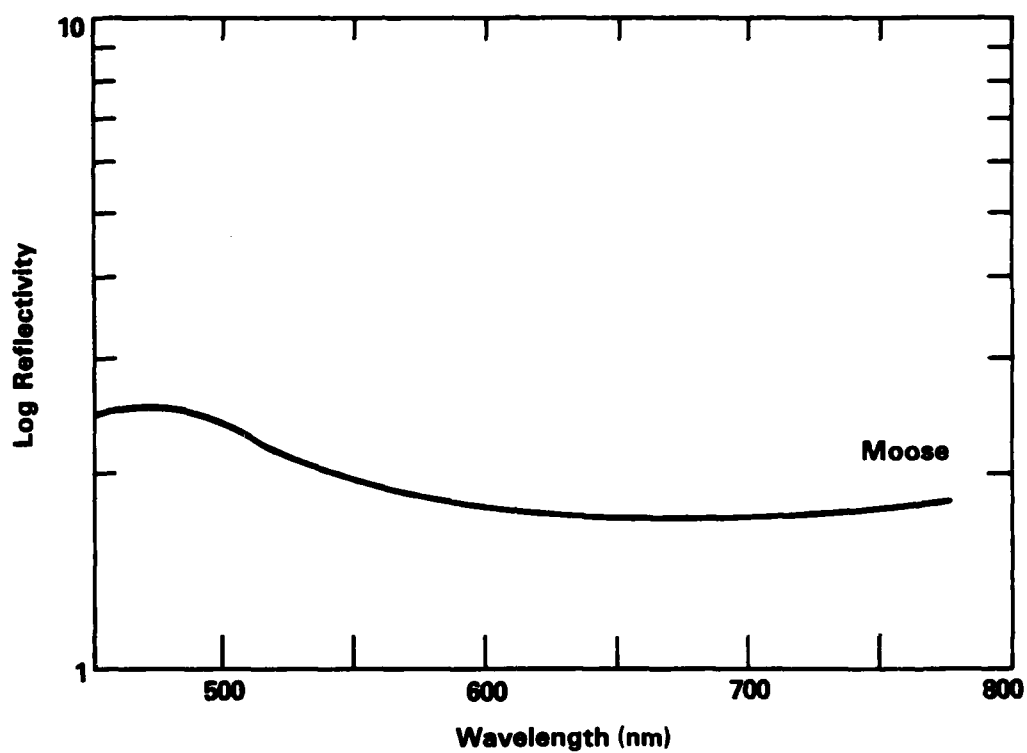




Fig. 4B

Moose

Fig. 5

Mule Deer

The mule deer retina is blue to blue-green over the tapetum and very dark elsewhere.

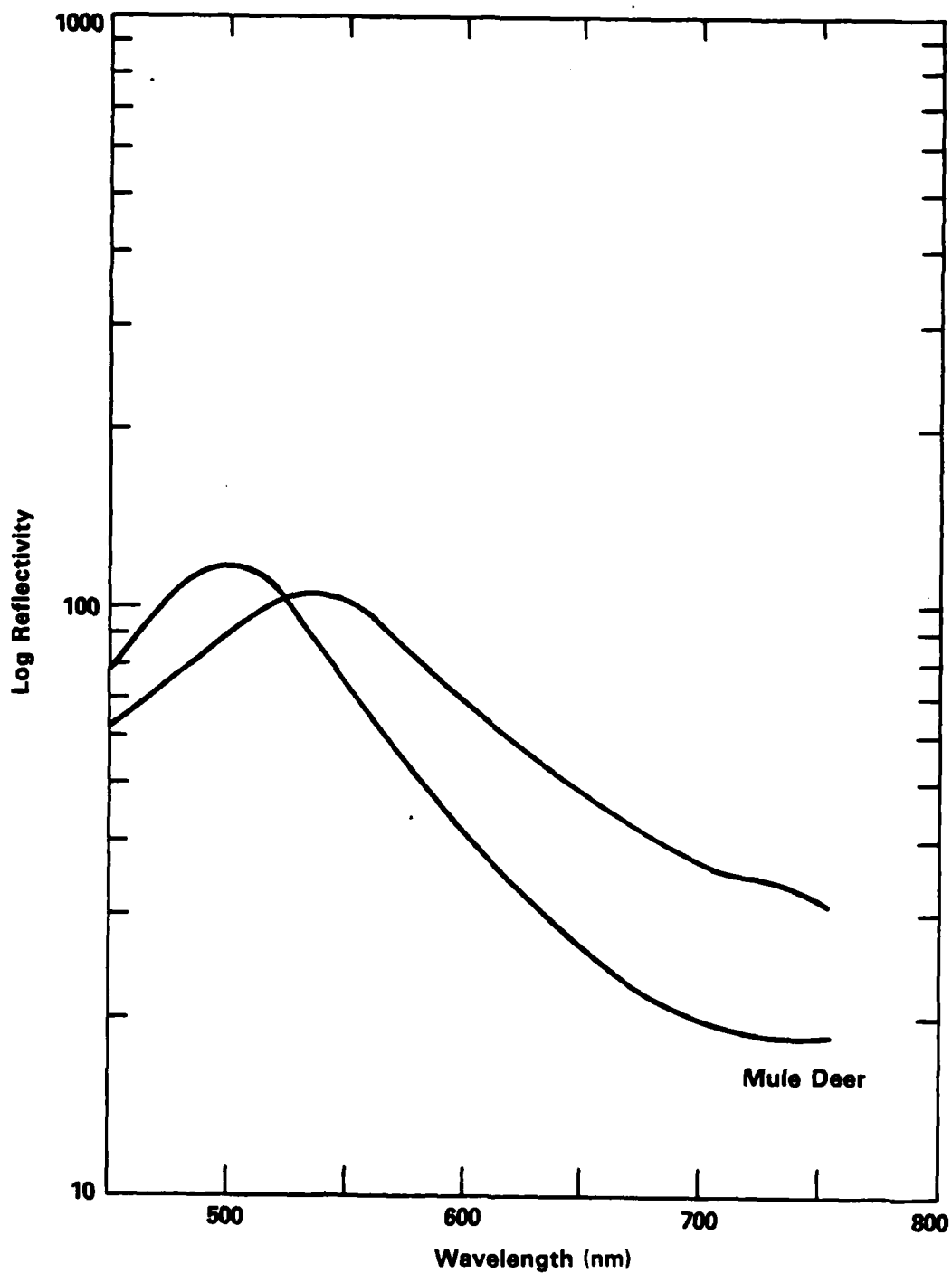




Fig. 5B

Mule Deer

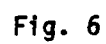


Fig. 6

Antelope

This antelope was very young, only several weeks old (see Fig. 21B). The blue cast of the tapetum is very evident in the spectral reflectance curve.

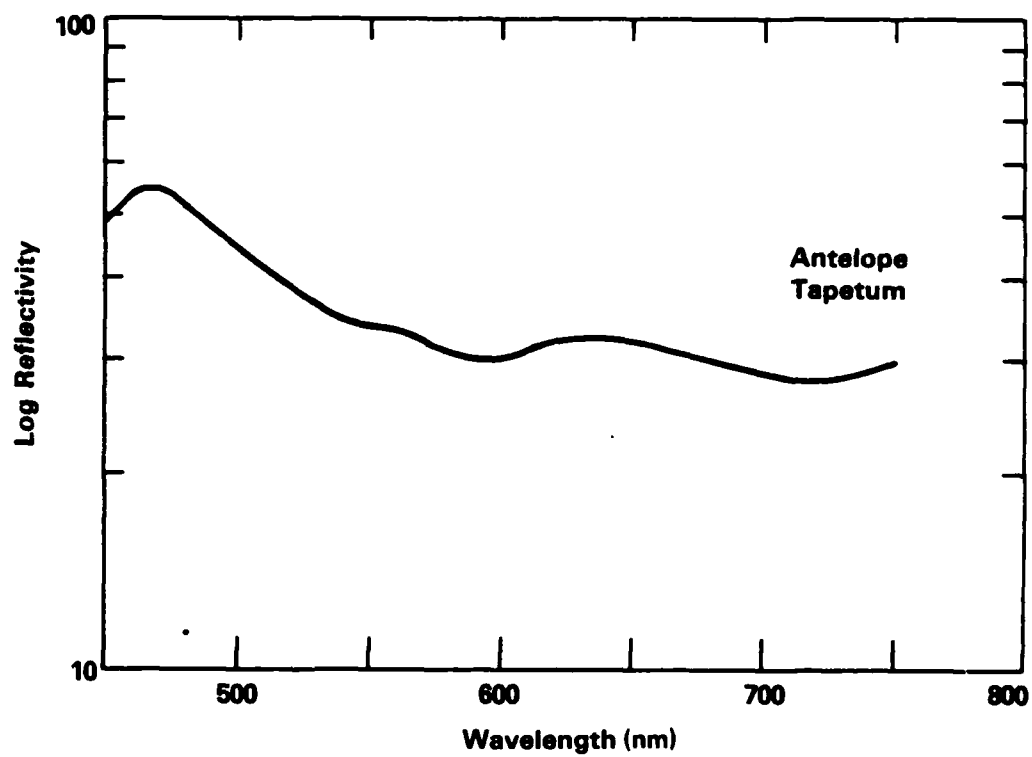




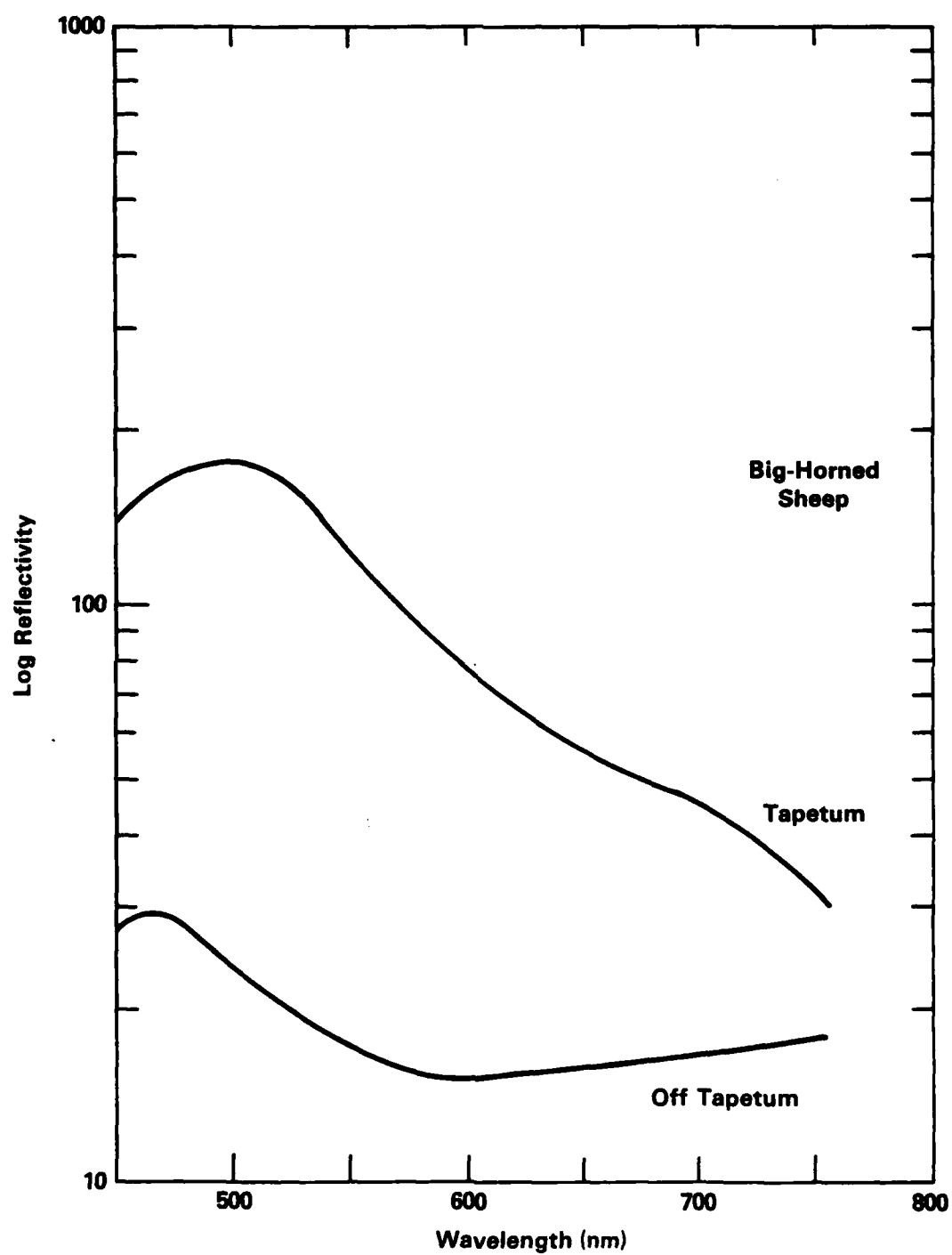
Fig. 68

Antelope

Fig. 7

Big-Horned Sheep

These retina are very highly reflecting, even in areas that are not covered with a tapetum. The tapetal color may be blue or blue-green.



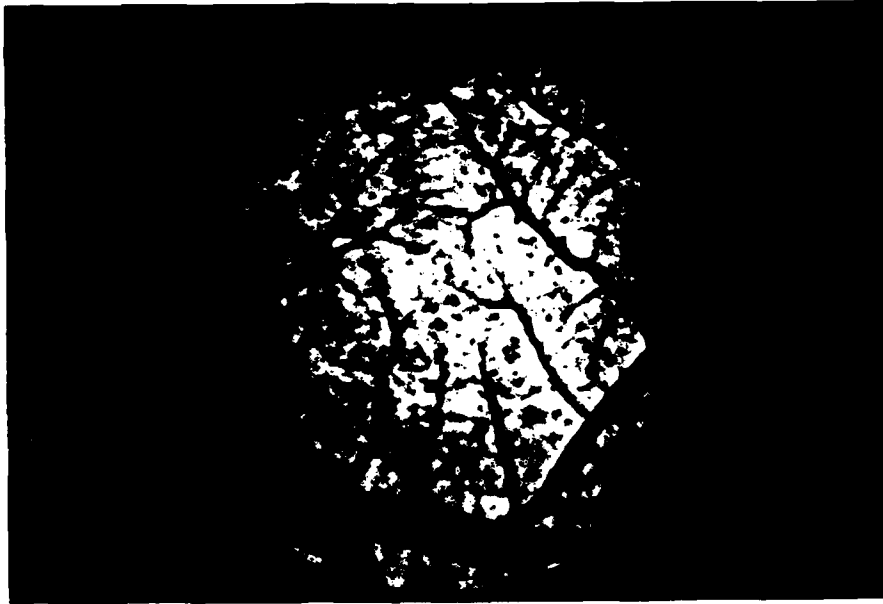


Fig. 7B

Big Horned Sheep



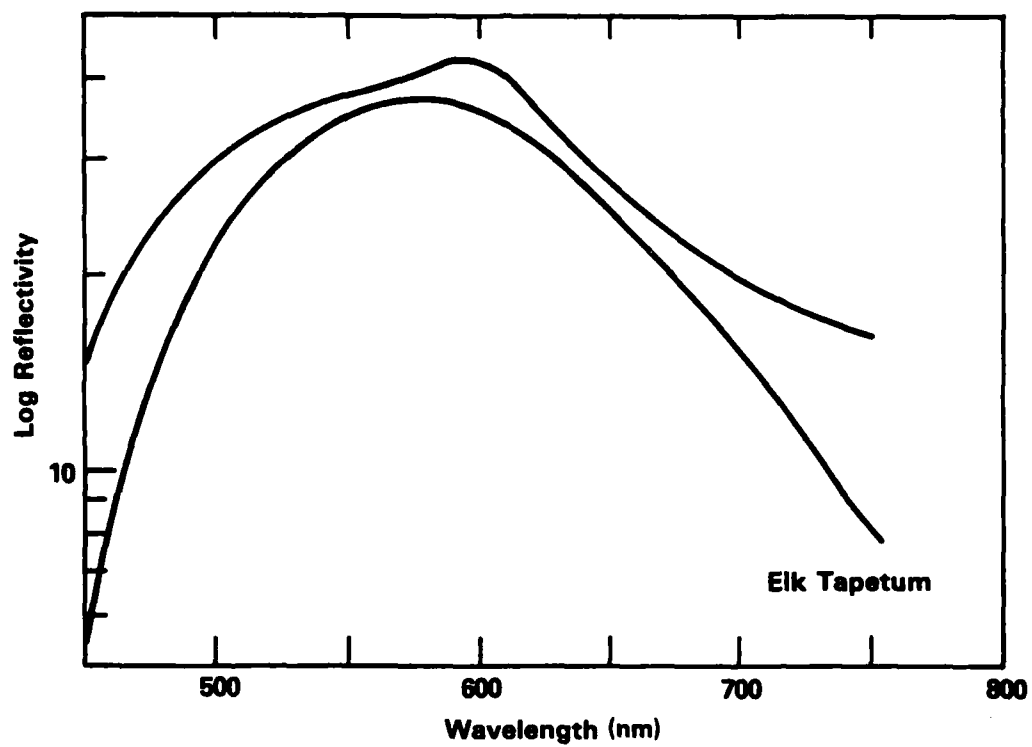
Fig. 7B

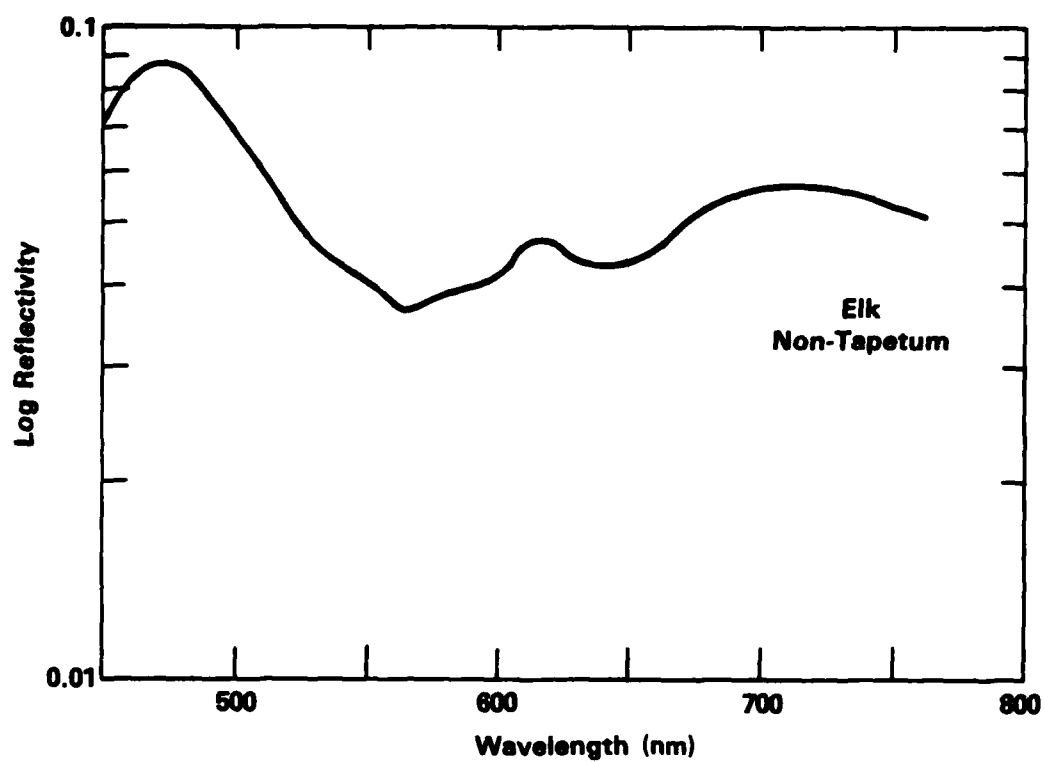
Big Horned Sheep

Fig. 8

Elk

The elk tapetum is very bright yellow, much different from most other animals.





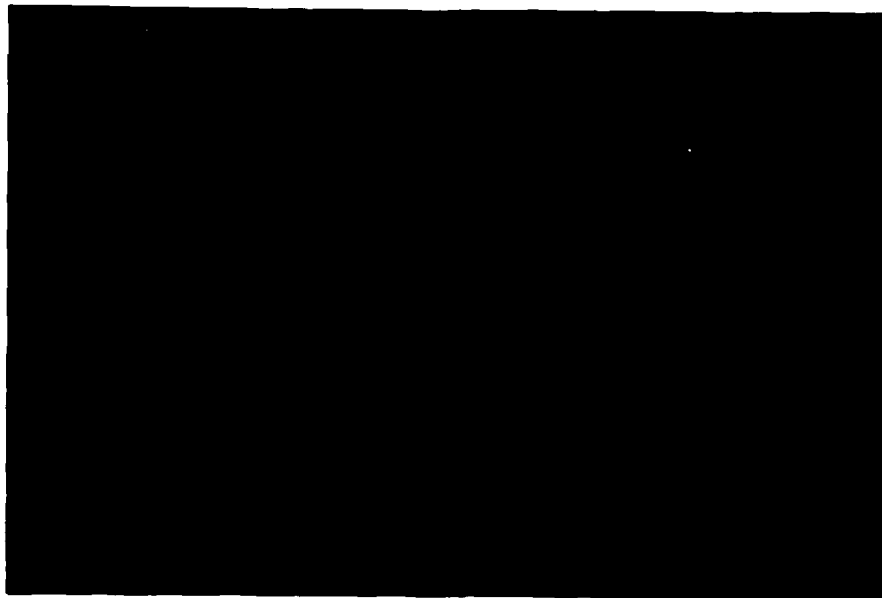


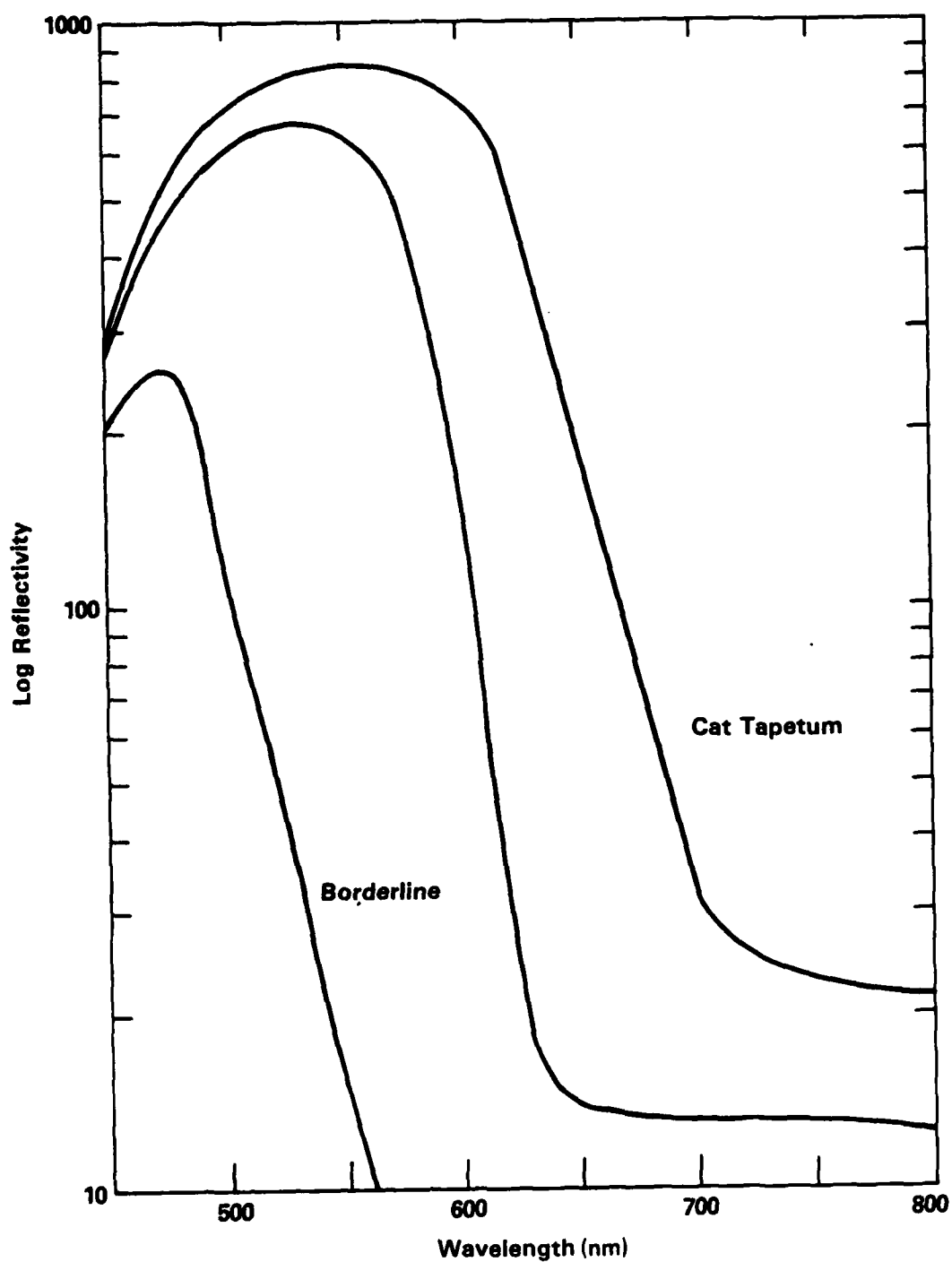
Fig. 8C

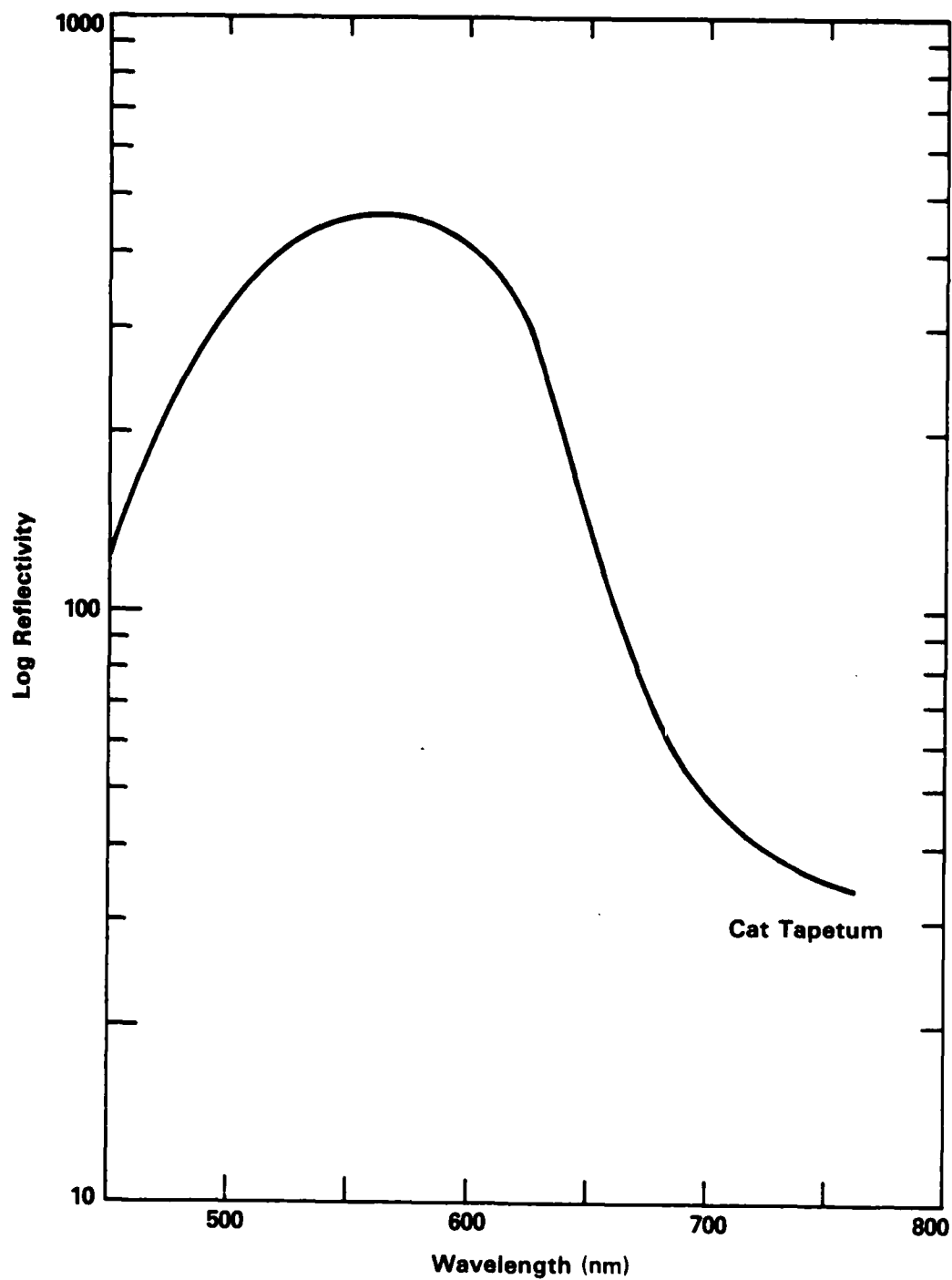
Elk

Fig. 9

Domestic Cat

The cat has the highest measured retinal reflectivity of any animal, probably due to its short focal length and the specular characteristics of the tapetum. The cat tapetum spectral curve is for cat #158, which is much yellower than #165. The cat tapetum near the optic disk is very blue.





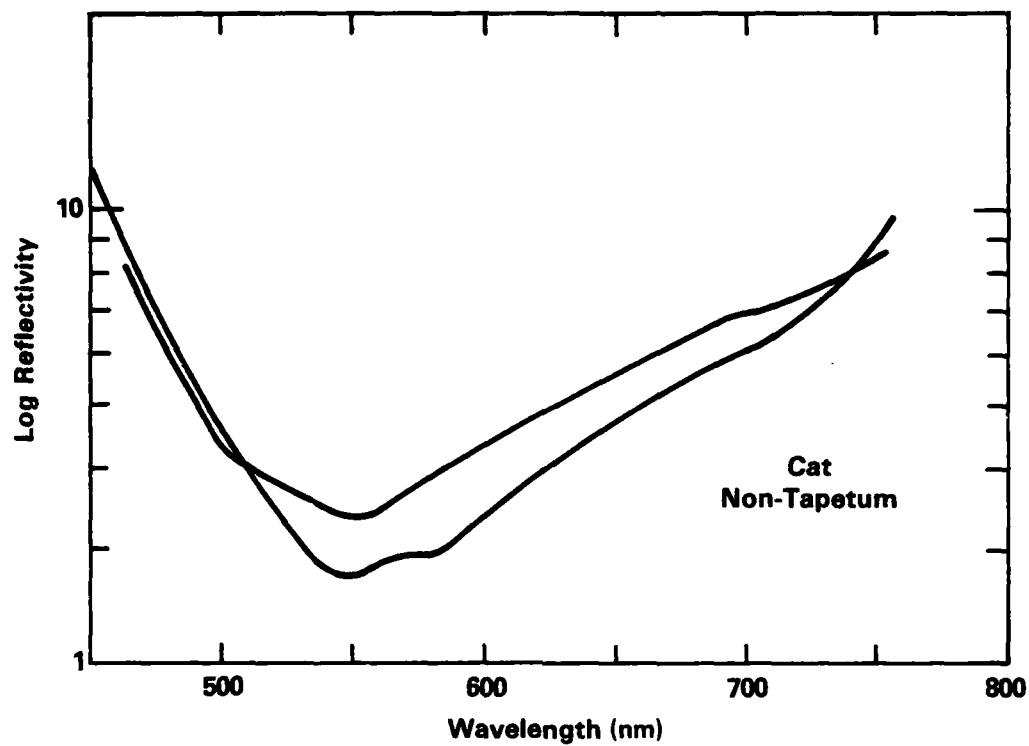




Fig. 9D

Cat #165

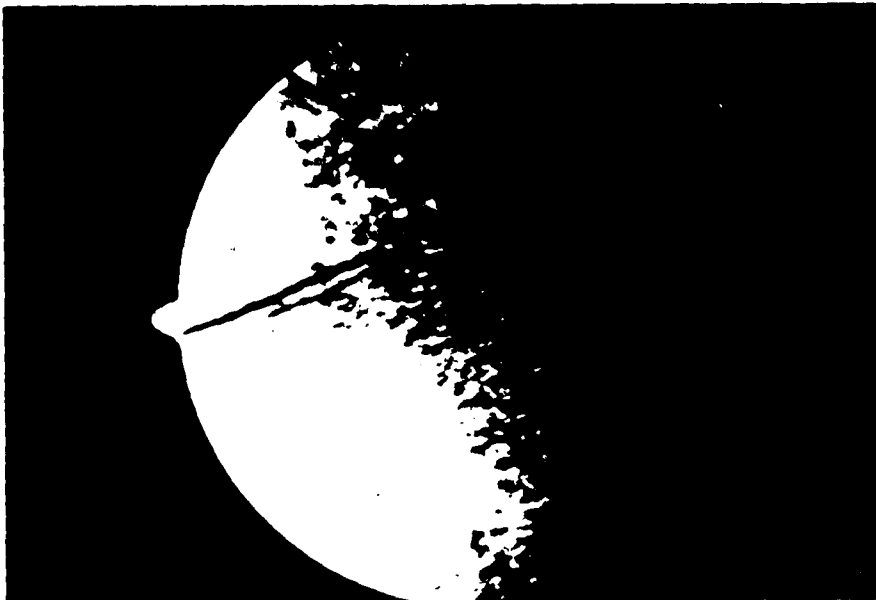
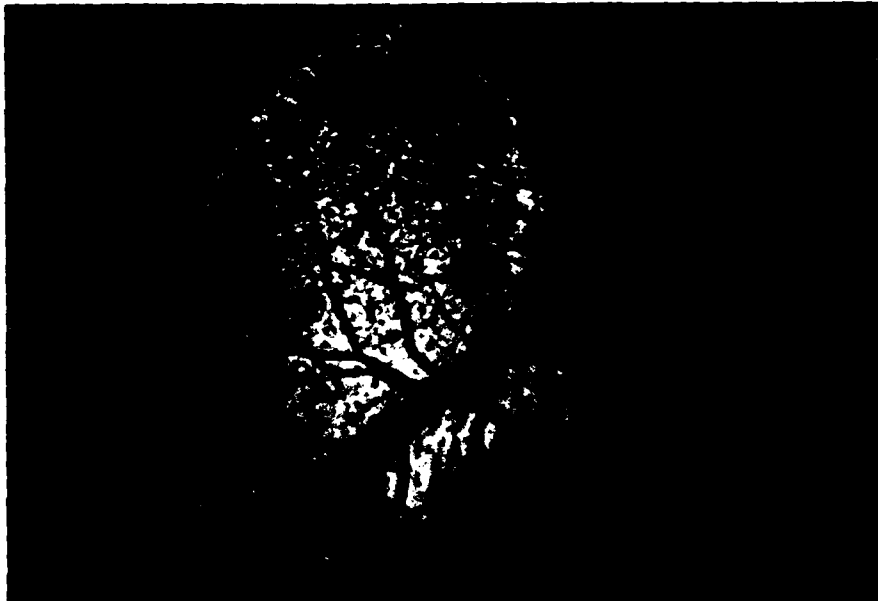


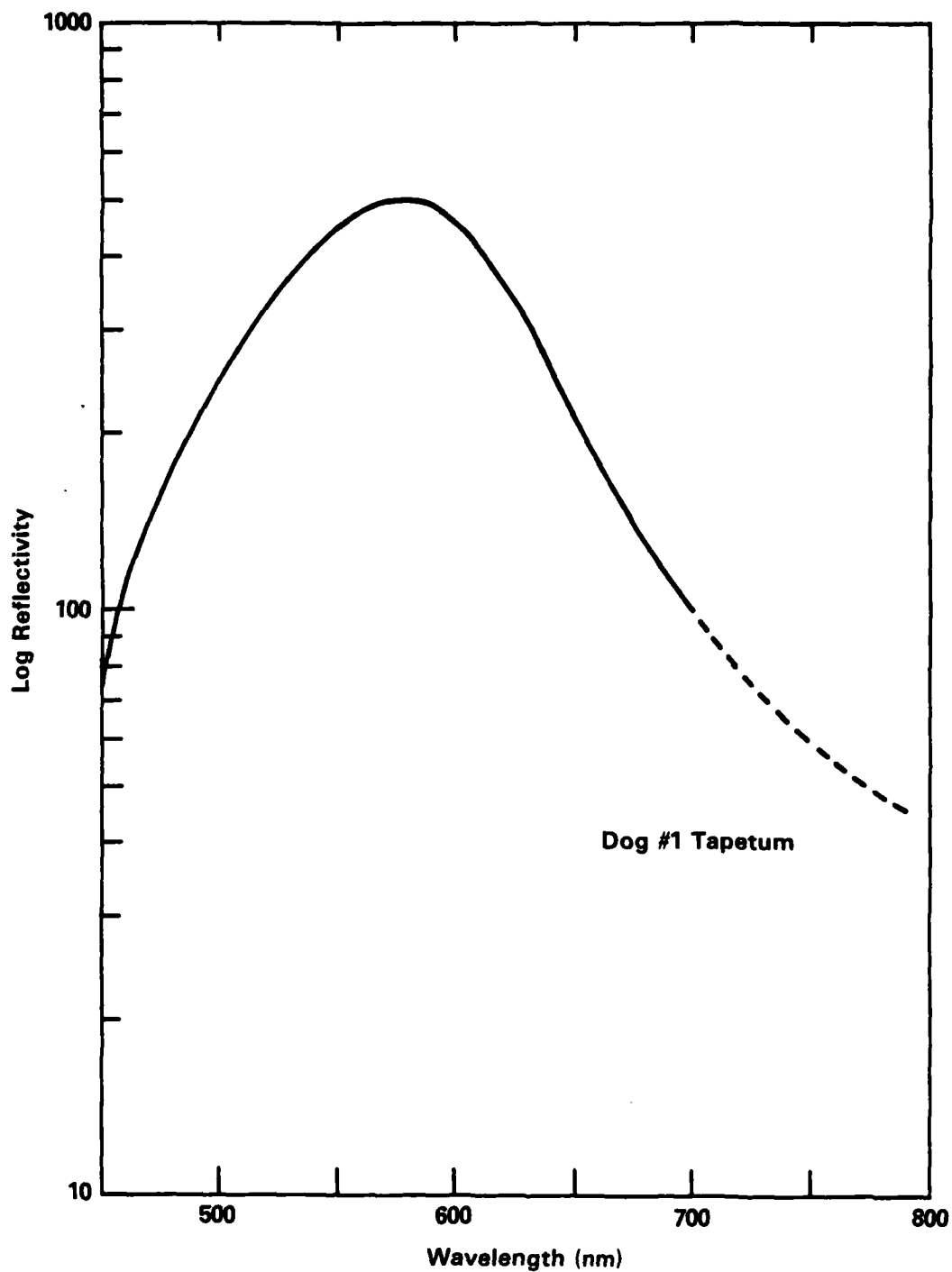
Fig. 9E

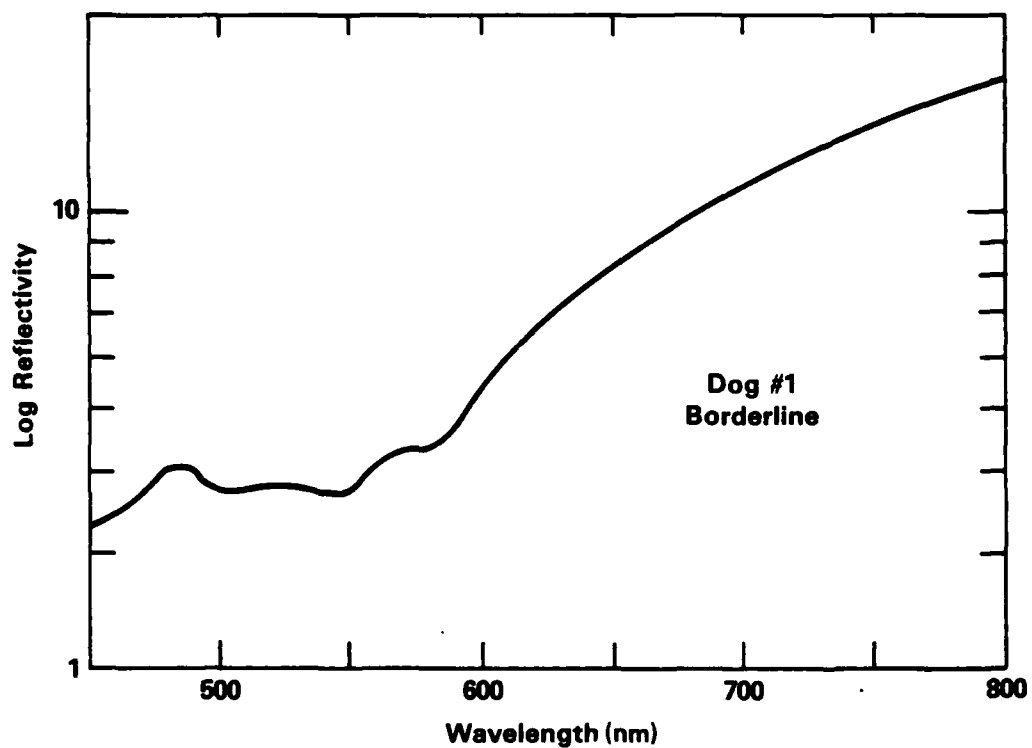
Cat =158

Fig. 10

Domestic Dog

Dog #1 is a beagle with a distinctive yellow tapetum.





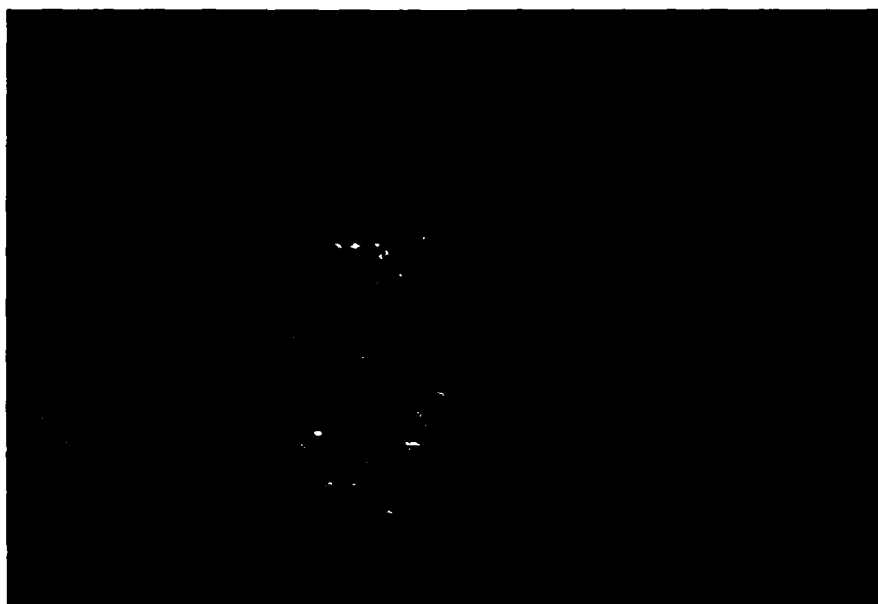
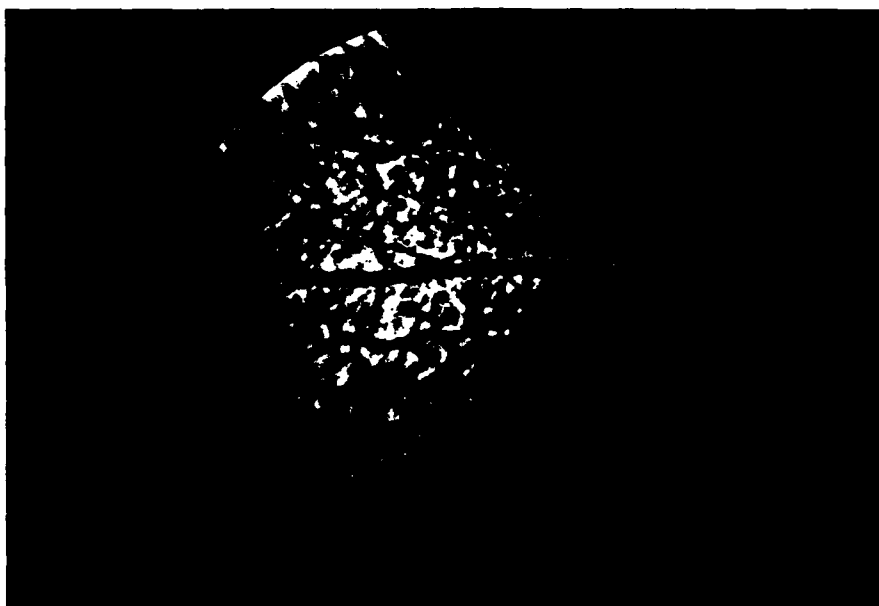
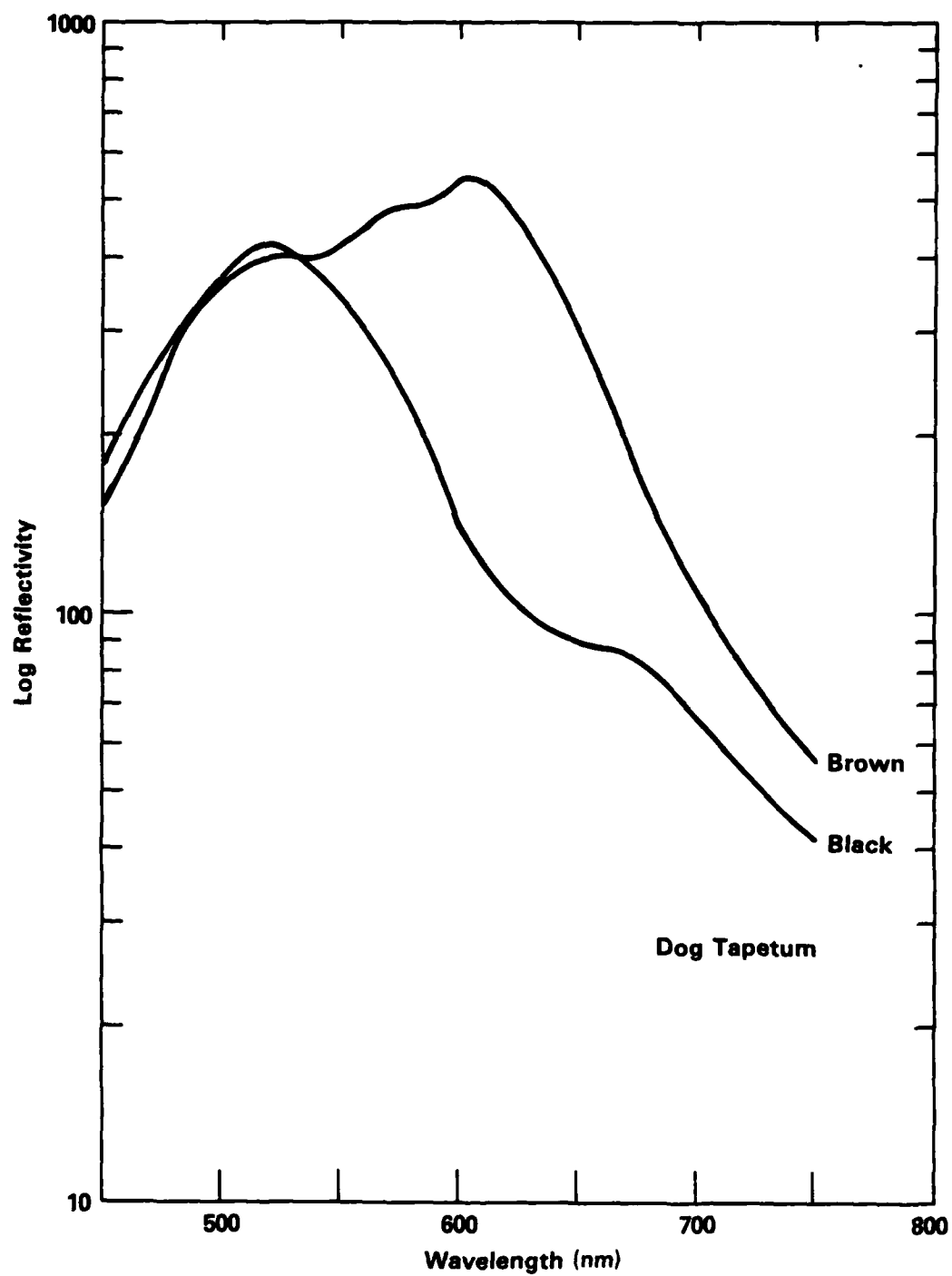


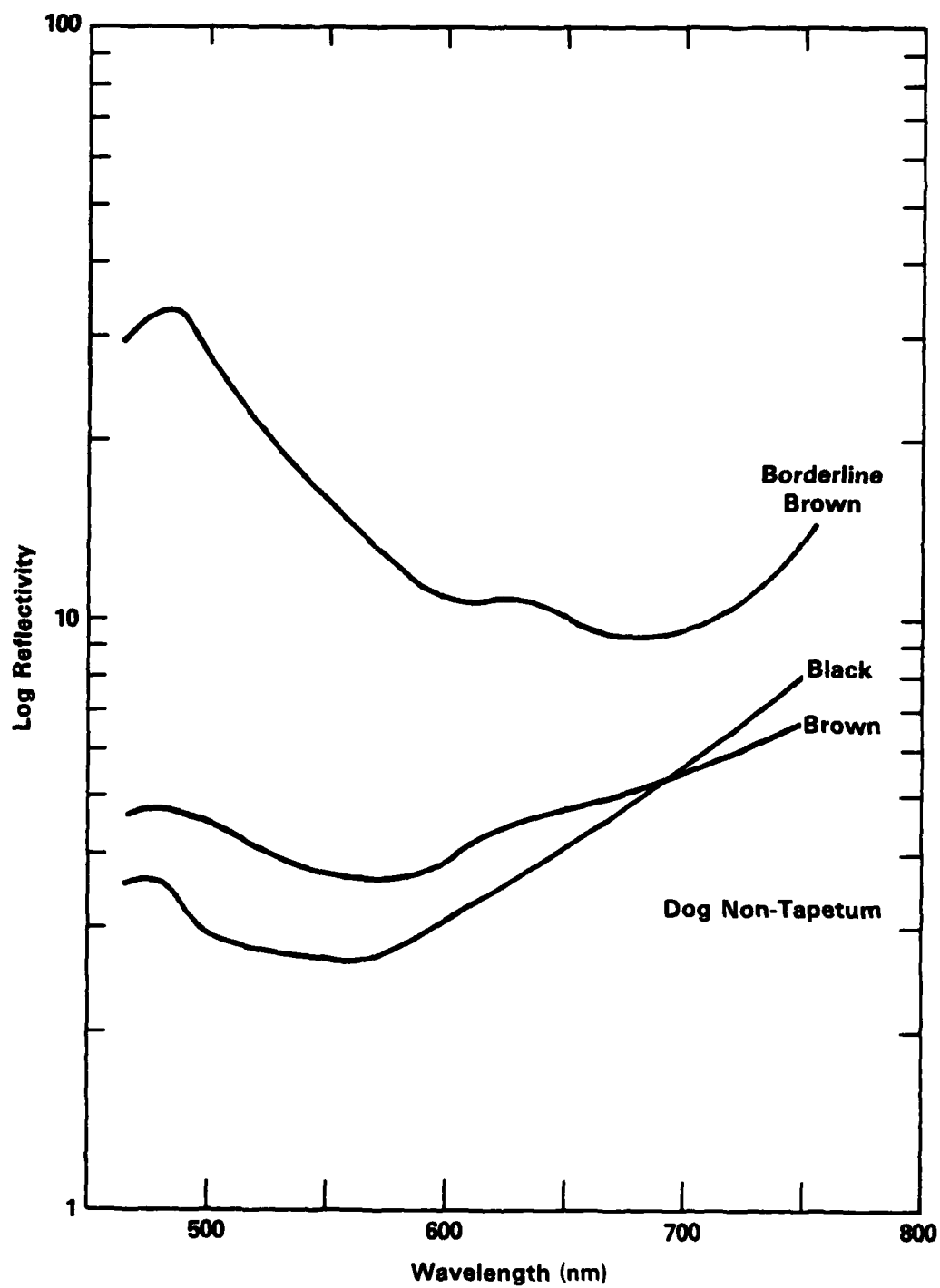
Fig. 10C

Dog #1

Fig. 11

Dogs #2 and #3 were short-haired mixed breed animals, one black and one brown in color. Their retinas are quite different, one blue and the other yellow. Note the difference in the spectral curves.





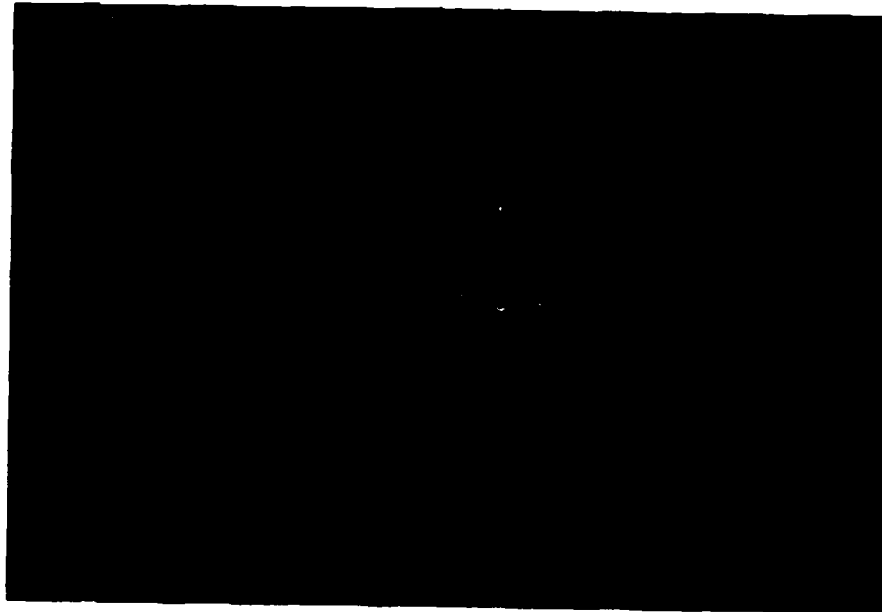


Fig. 11C

Dog Brown



Fig. 11D

Dog Black

Fig. 12

Farm Goat

The blue tapetum is seen in both the photograph and the spectral curves.

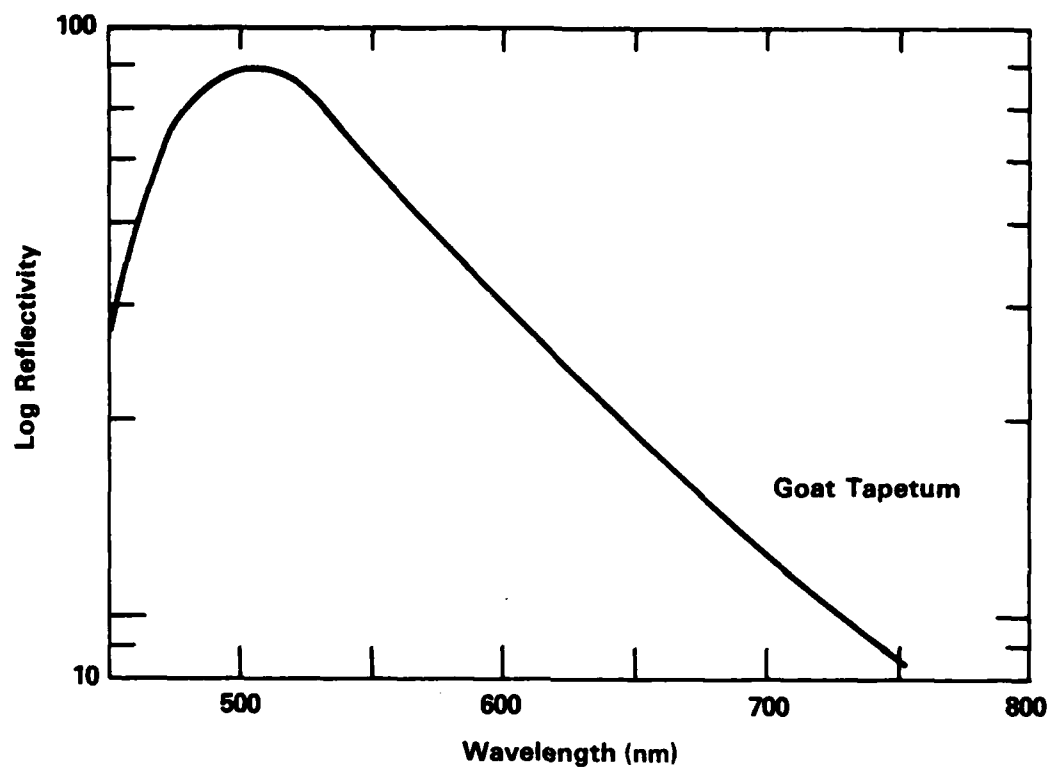




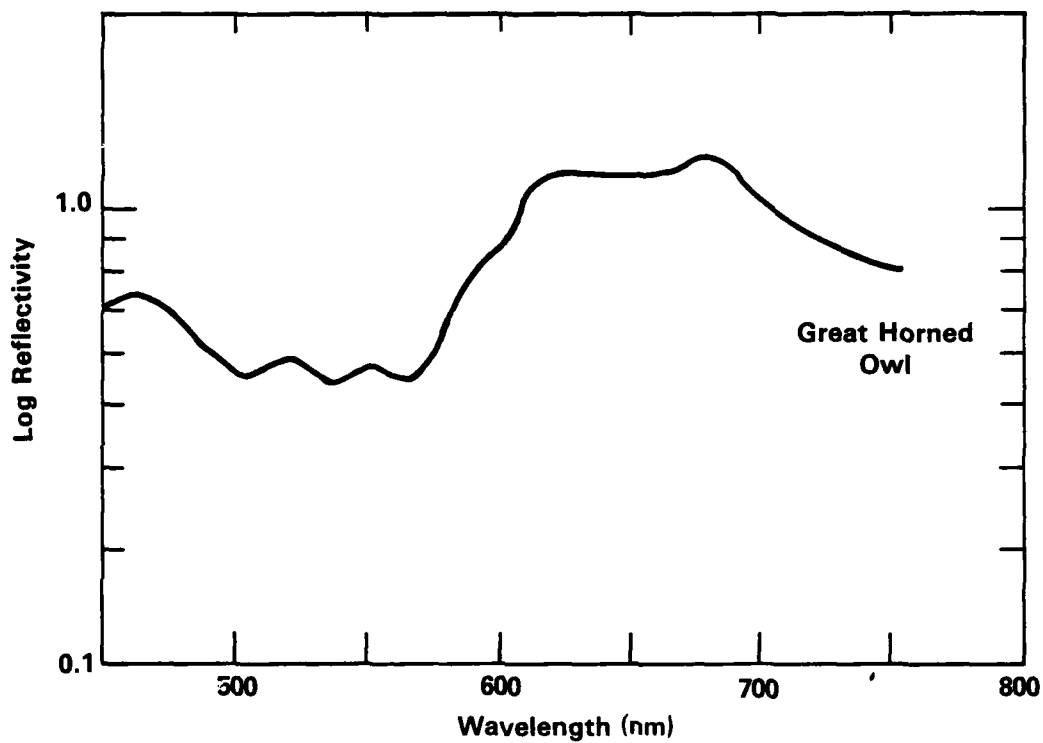
Fig. 12B

Goat

Fig. 13

Great Horned Owl

The owl retina is very dark and red. In the photograph the pecten can be easily seen. This is common to many bird species.



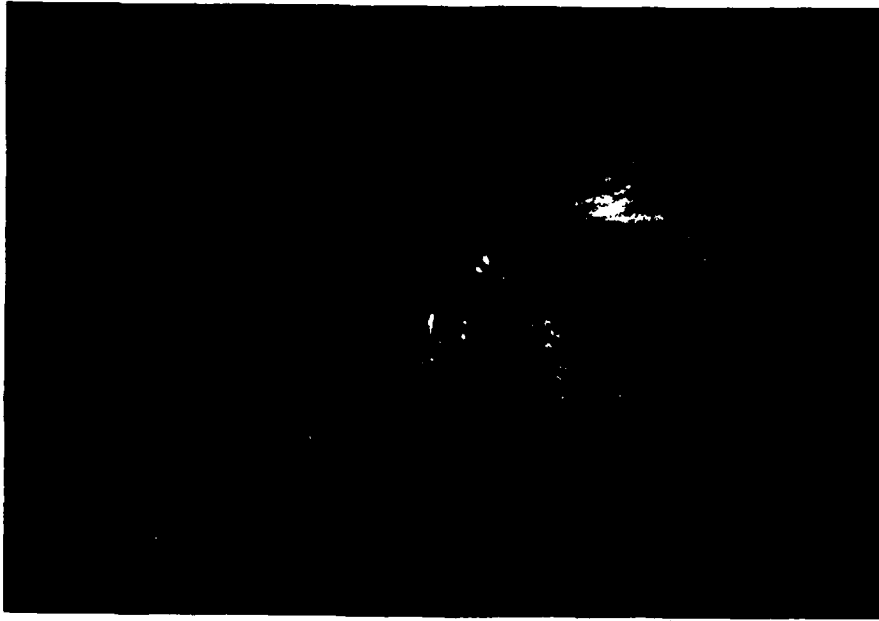


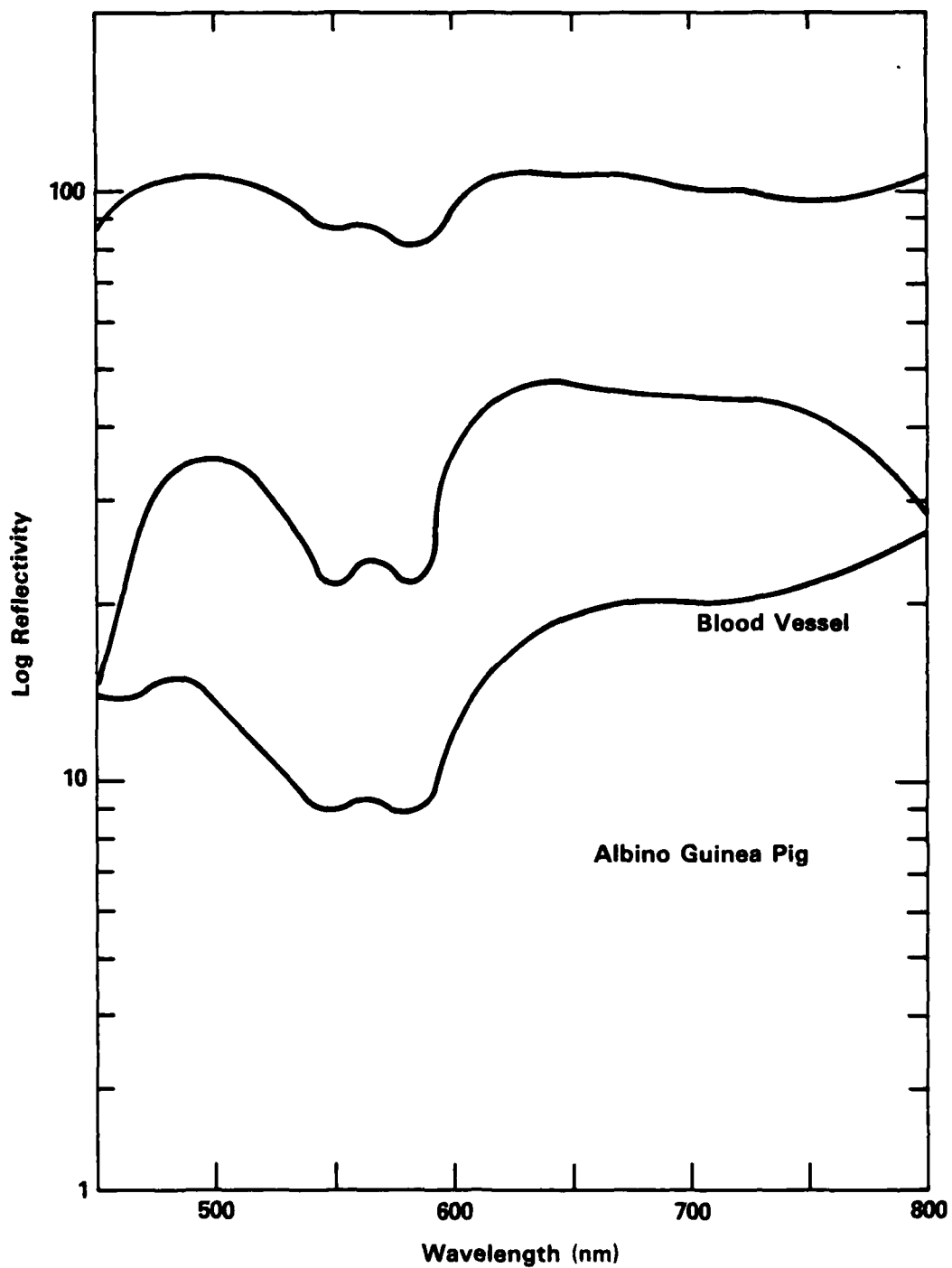
Fig. 13B

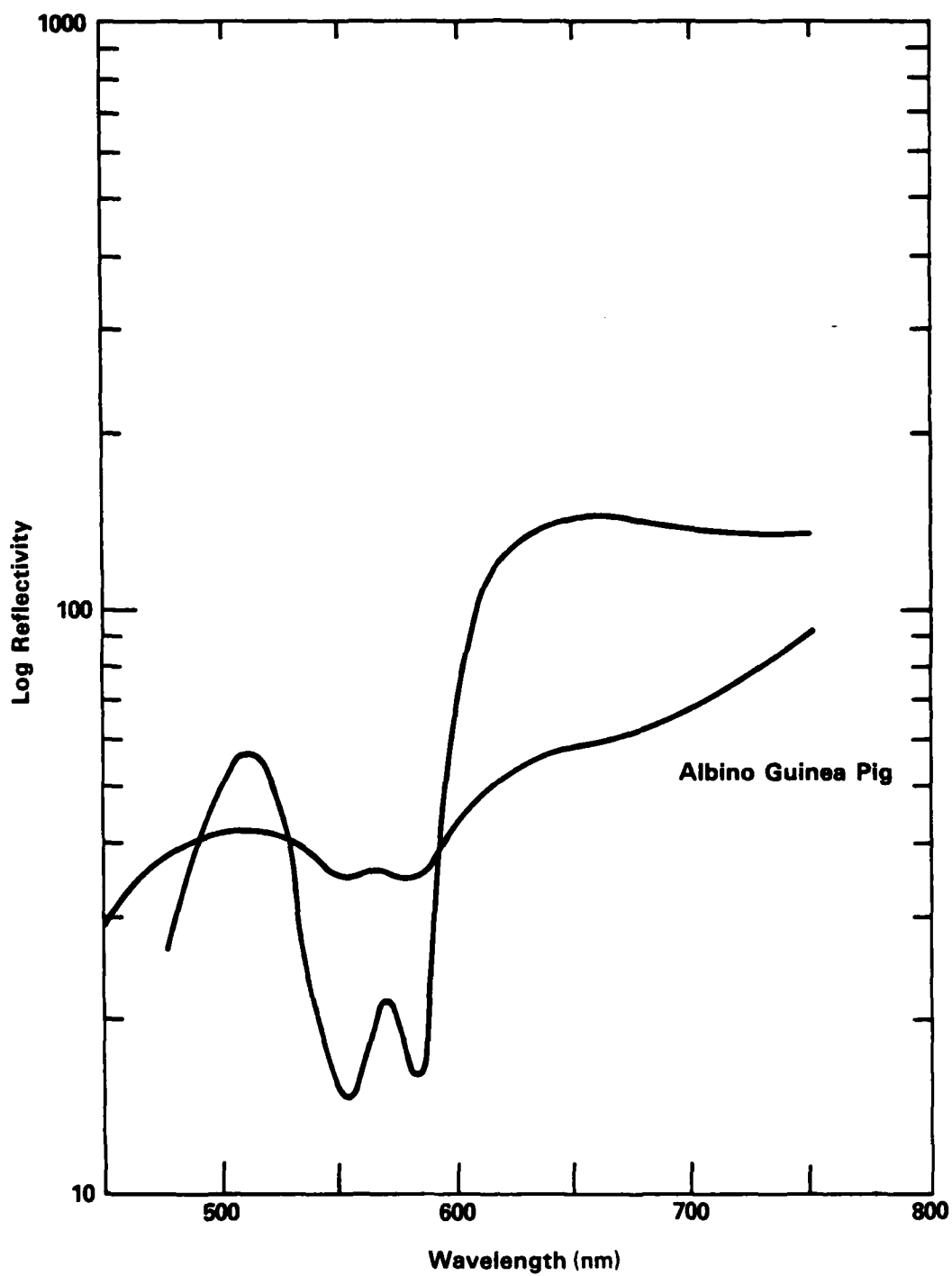
Great Horned Owl

Fig. 14

Albino Guinea Pig

Albino guinea pigs and albino rabbits have very similar appearing retinas when the disk is not in the field of view.





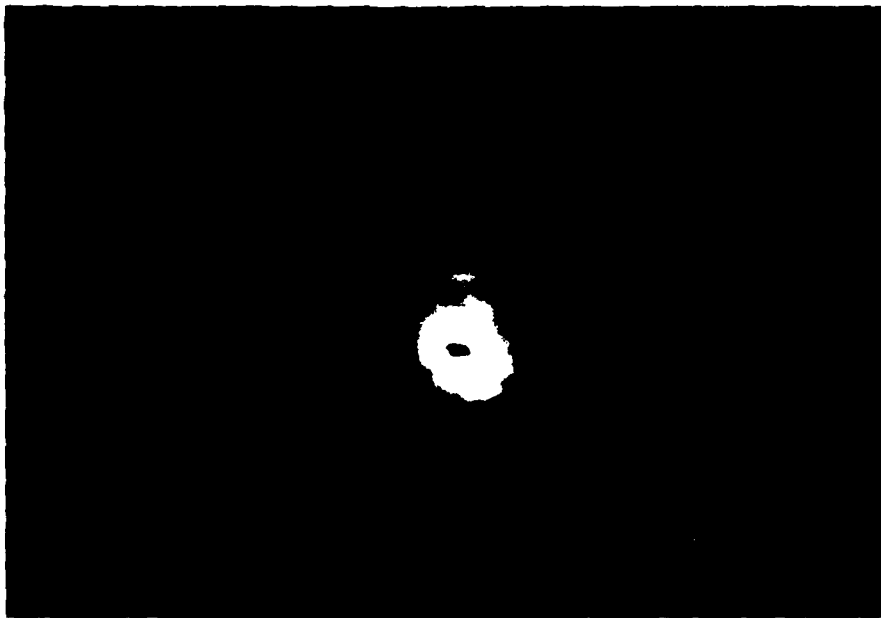


Fig. 14C

Albino Guinea Pig

Fig. 15

Pigmented Guinea Pigs

The pigmented guinea pig has such a dark retina and small pupil that photographs could not be obtained.

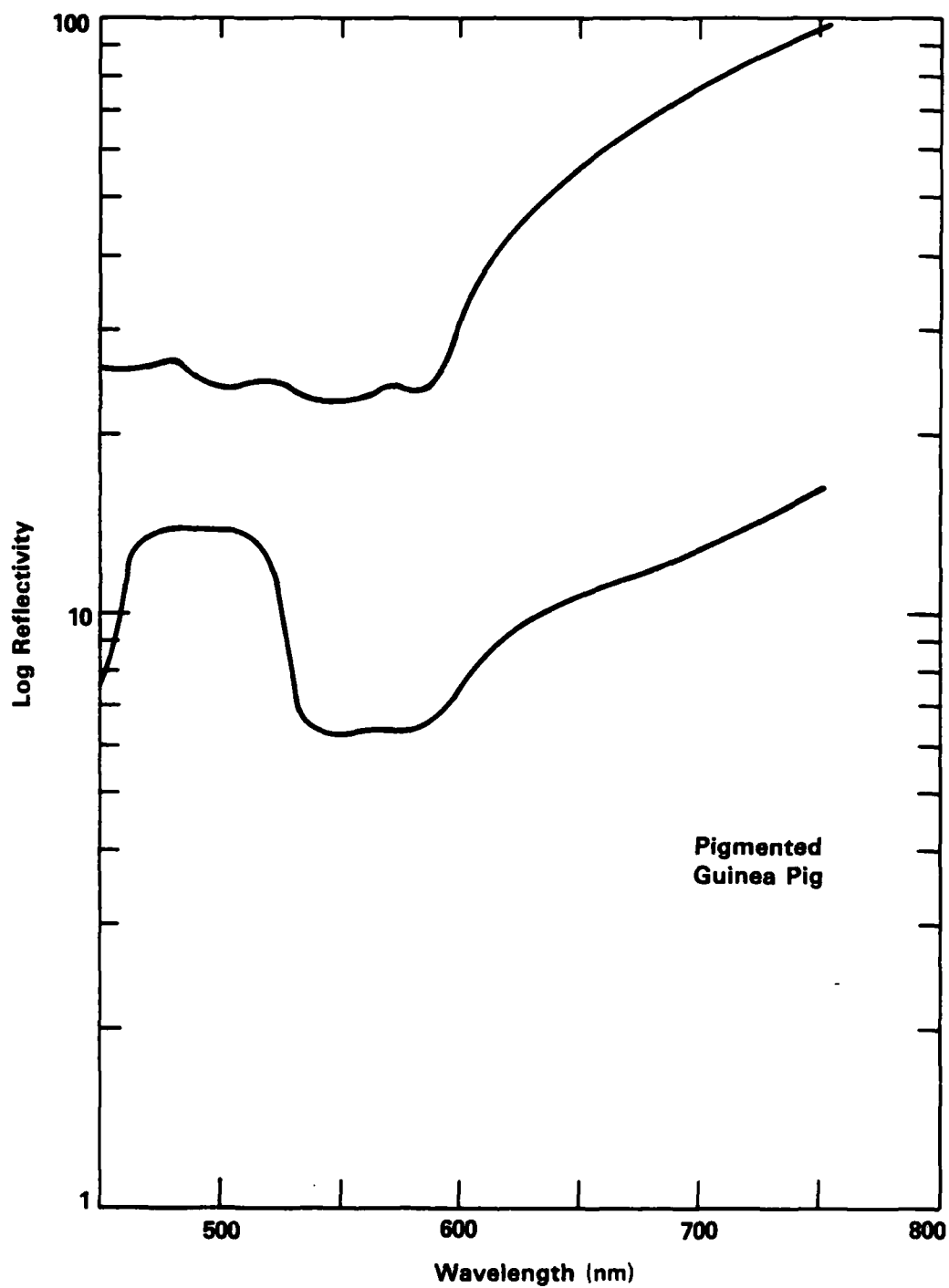
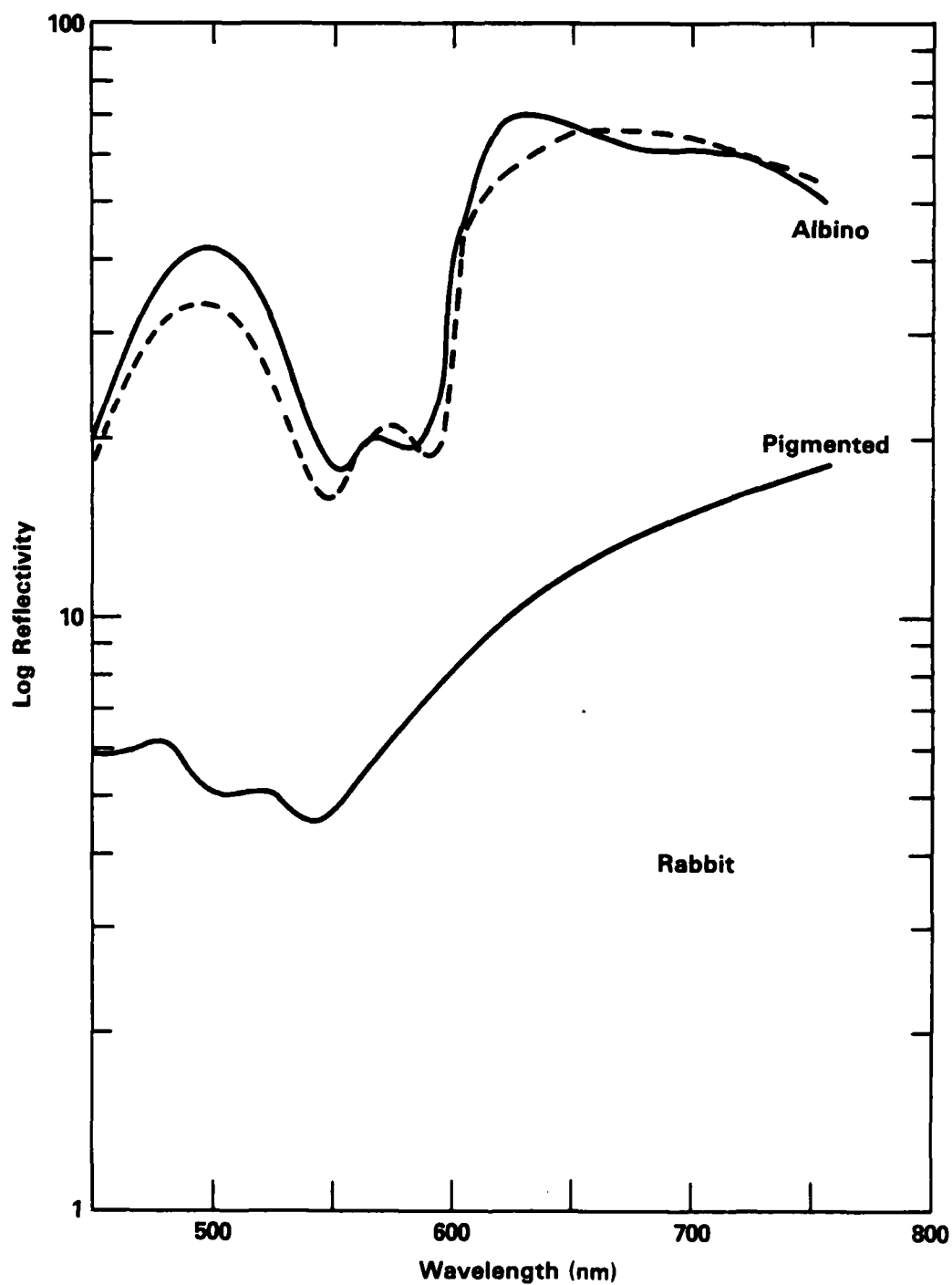
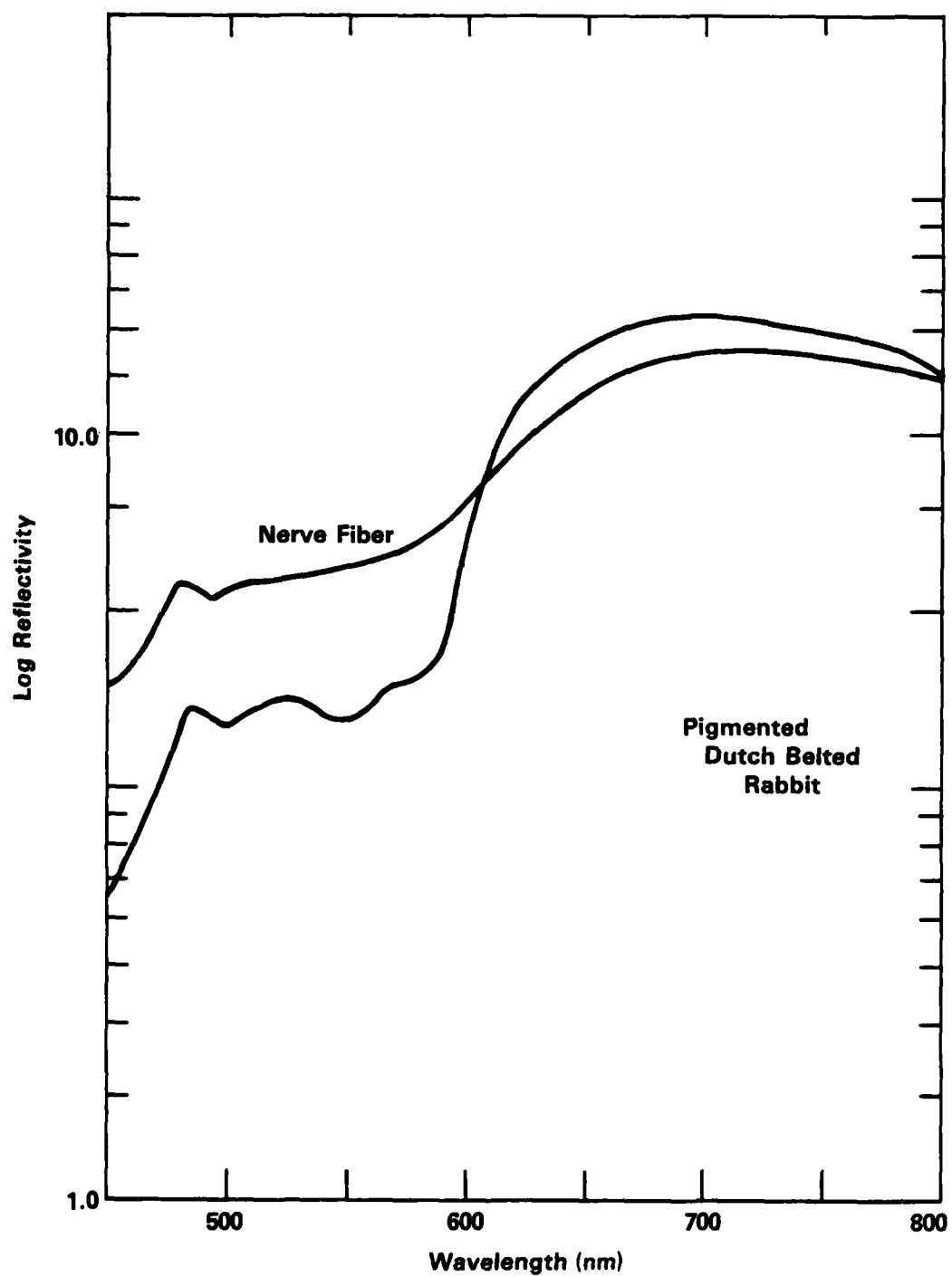


Fig. 16

Rabbits

In the albino rabbit spectral curves the absorption of blood is easily seen.





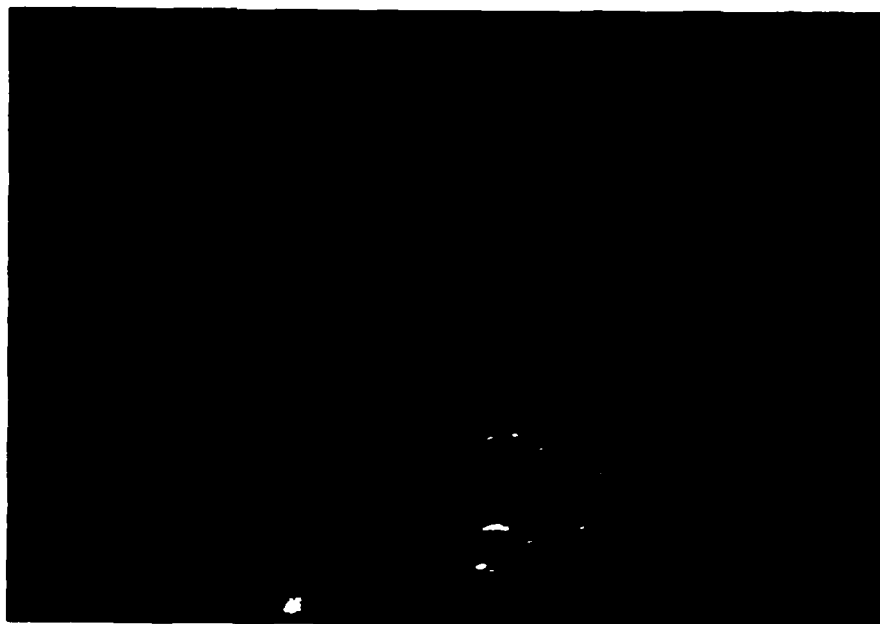
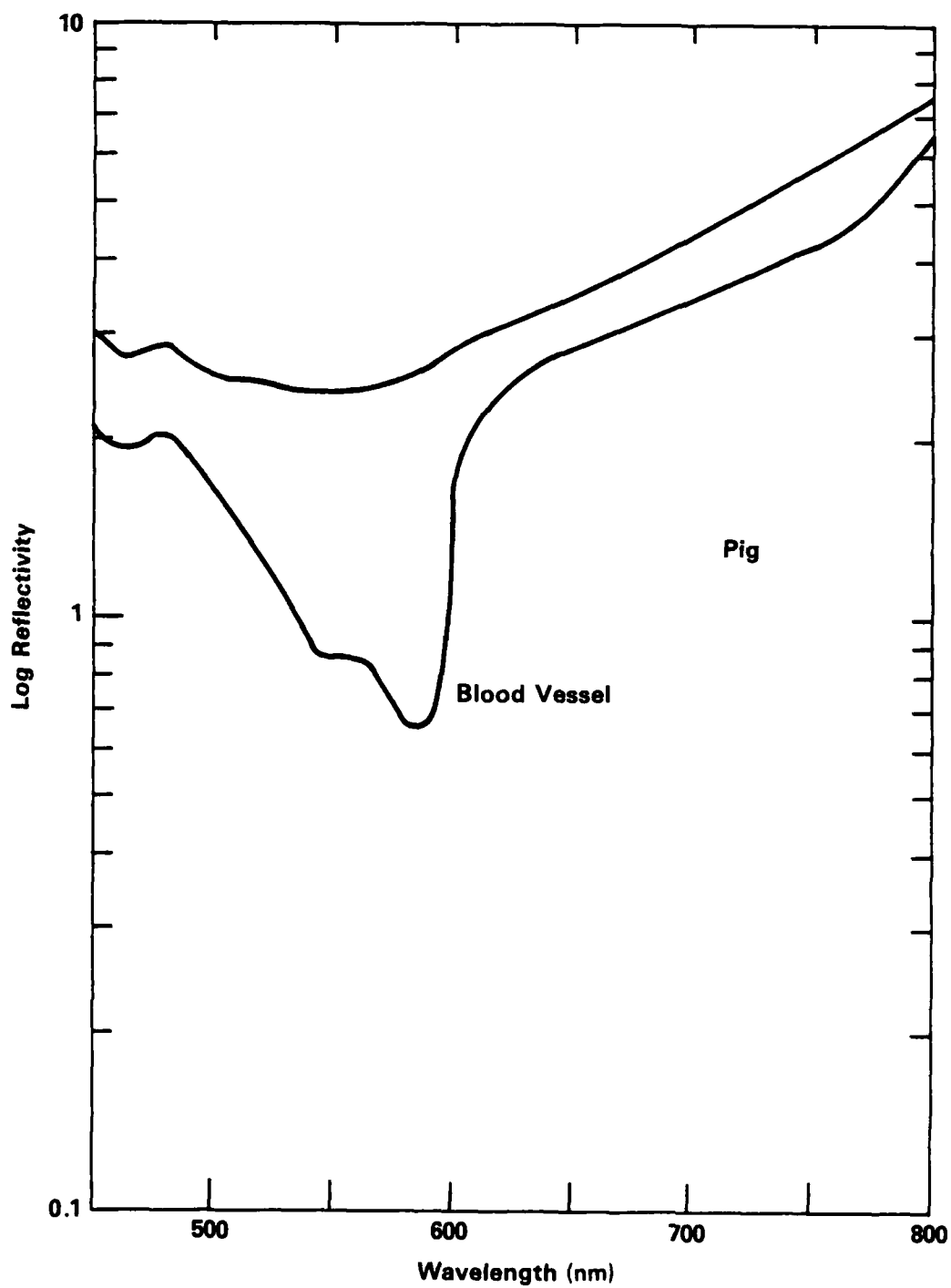


Fig. 16C

Fig. 17

Farm Pig

The pig retina looks much like a human or monkey retina except for a greyish tinge and the absence of a well-defined macula.





Pig "Henry"



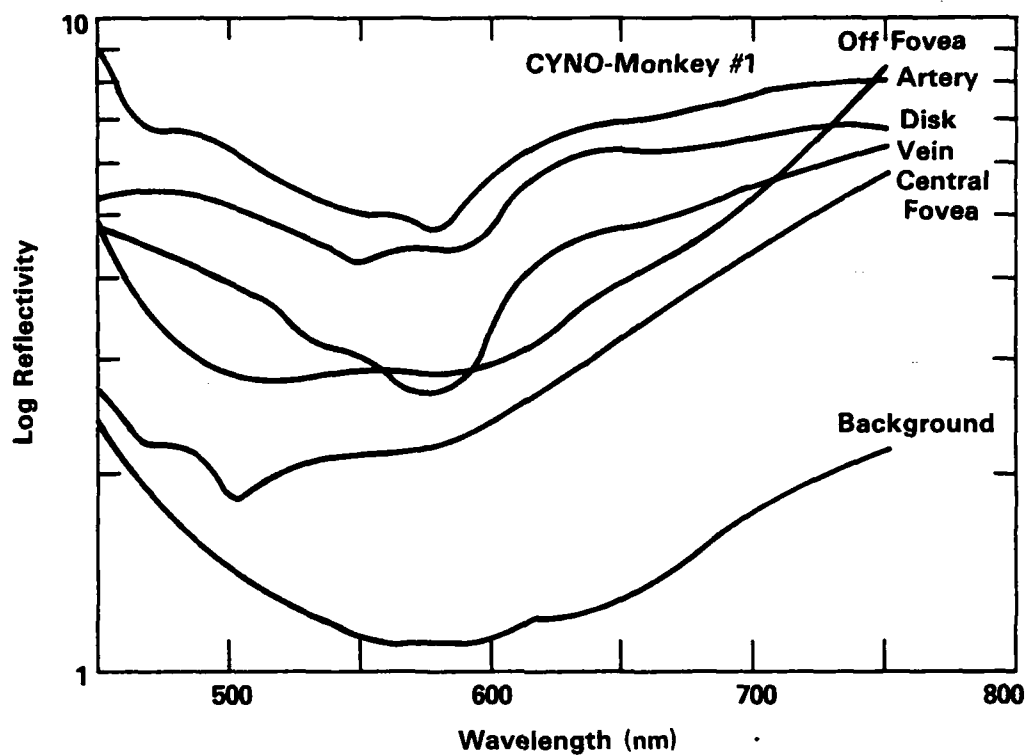
Pig "Clyde"

Fig. 18

Cynomolgus Monkey

The areas indicated by dots were measured. The artery spectral curve is very high due to a reflective highlight.

Fig. 18C, the central fovea of two different animals, shows evidence of a yellow absorbing pigment. These two animals are almost identical at the central fovea but (see Fig. 16D) not over the whole retina.



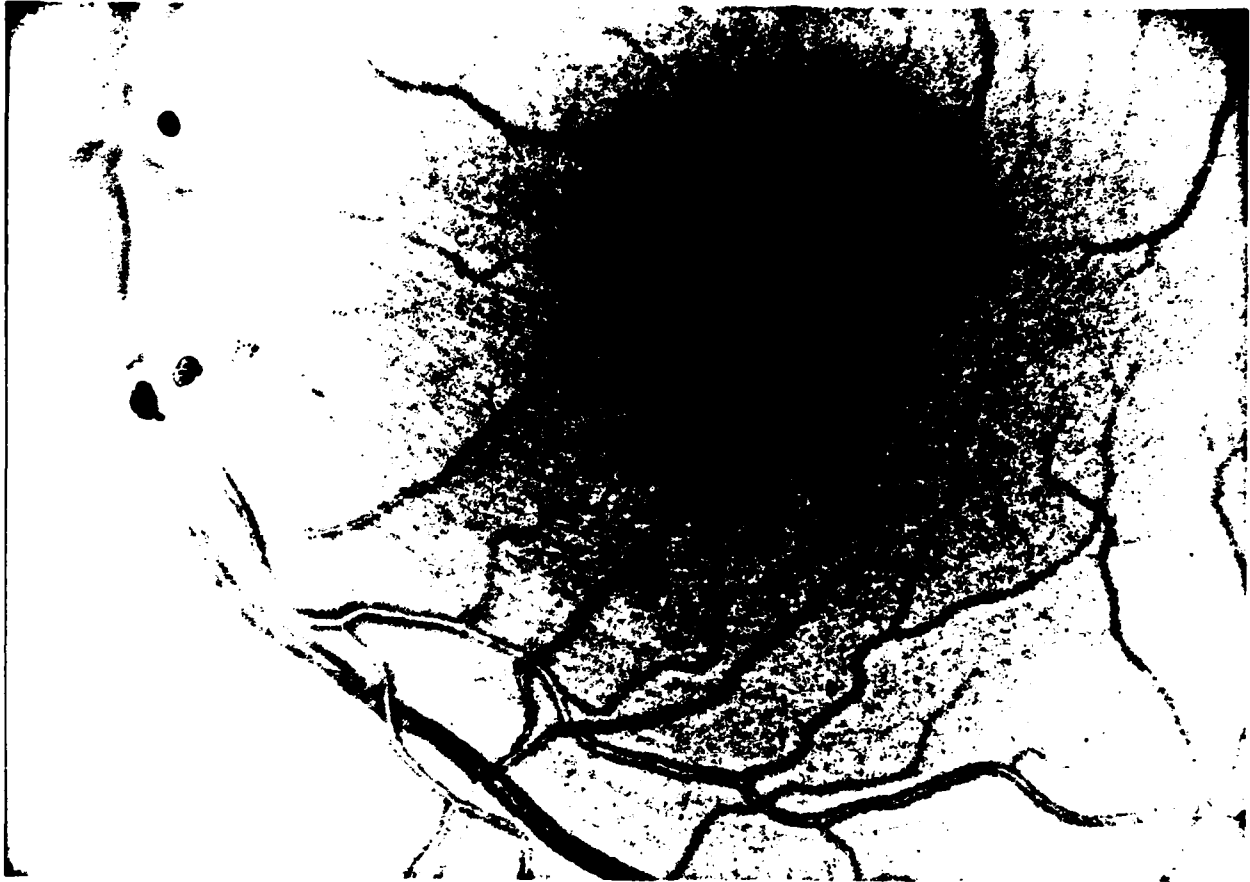
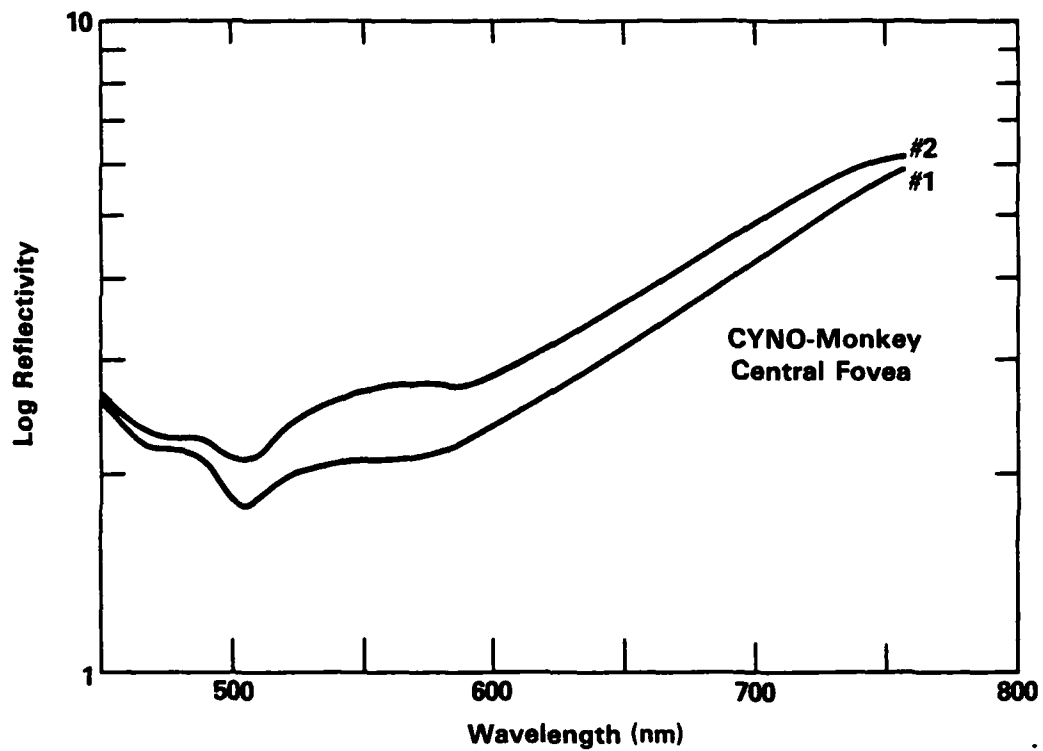
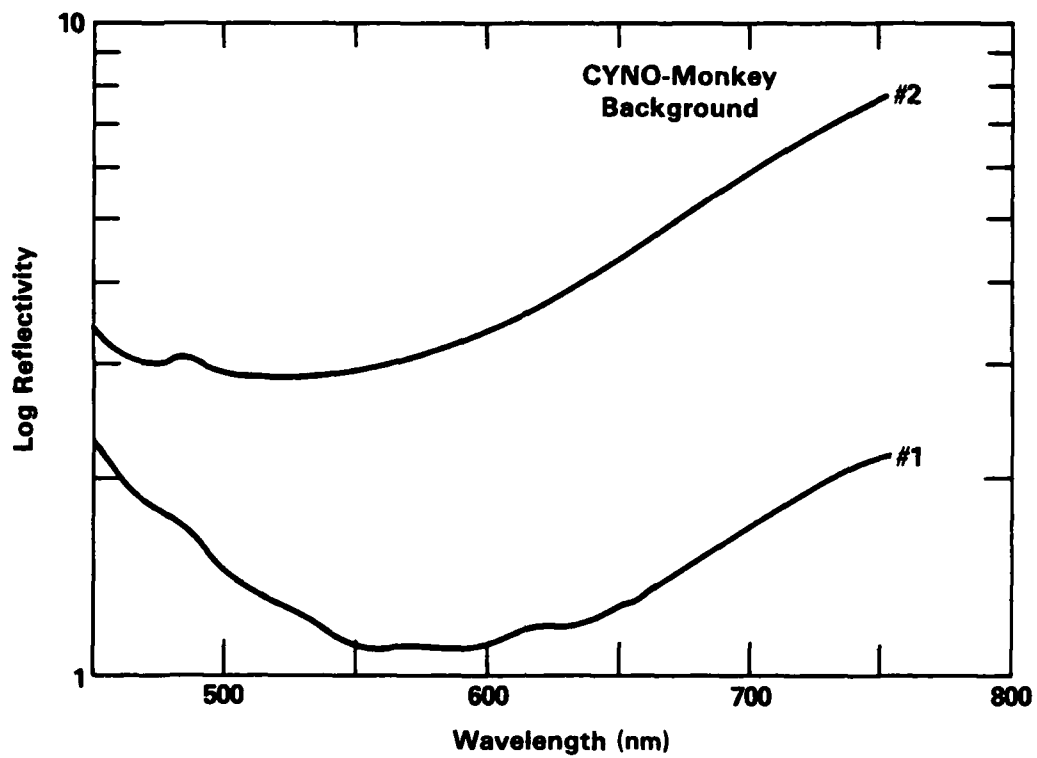


Fig. 18B





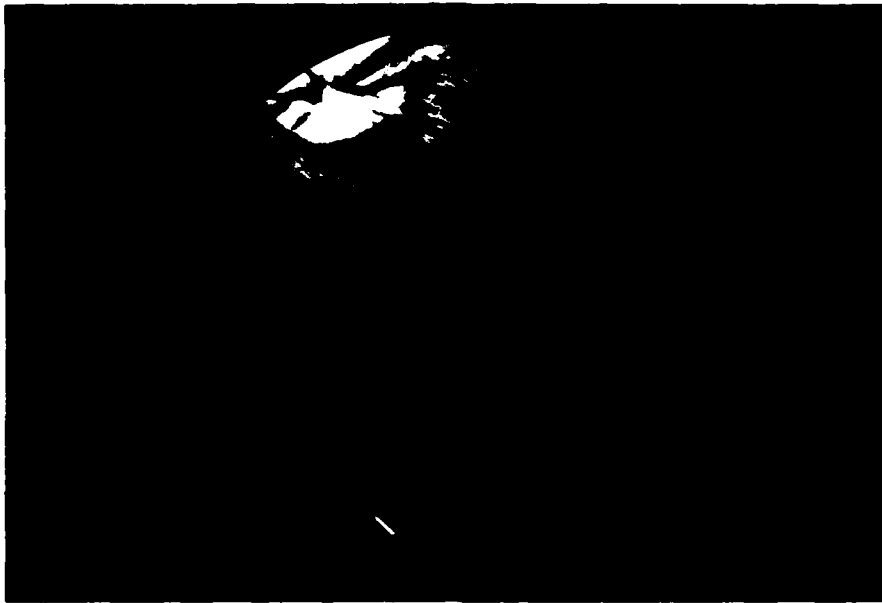


Fig. 18E

Monkey #2



Fig. 19

Fig. 20

A Mule Deer

B Moose

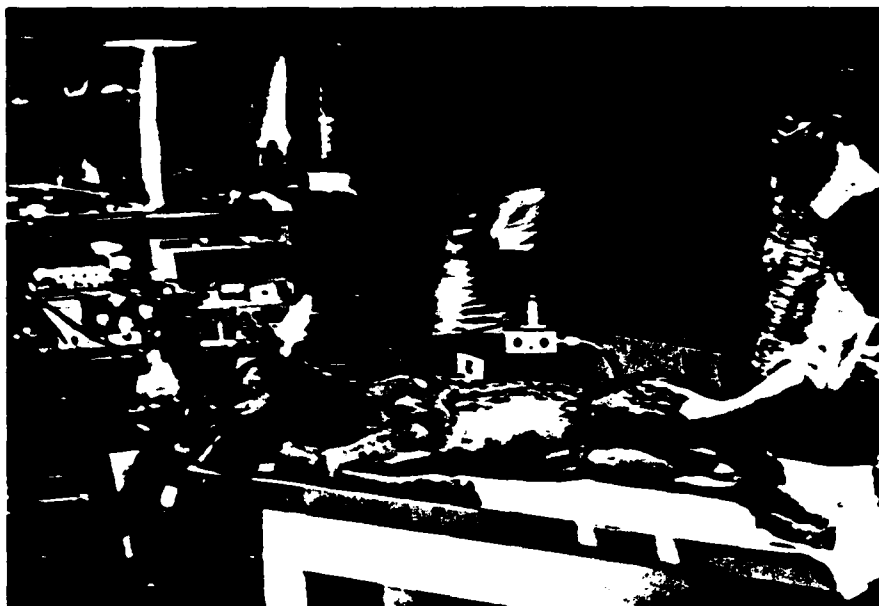


Fig. 20

Fig. 21

A Moose

B Antelope



Fig. 21

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